MEDICAL LIBRARY AUG 16 1938

VOLUME 26 NUMBER 2

ARCHIVES OF PATHOLOGY

EDITORIAL BOARD

LUDVIG HEKTOEN, Chicago, Chief Editor

JAMES EWING, New York
OSCAR T. SCHULTZ, Evanston, Ill.
S. B. WOLBACH, Boston
ALFRED STENGEL, Philadelphia
FRANK B. MALLORY, Portland, Ore.

AUGUST 1938

PUBLISHED MONTHLY BY AMERICAN MEDICAL ASSOCIATION, 535 NORTH DEARBORN STREET, CHICAGO, ILLINOIS. ANNUAL SUBSCRIPTION, $8.00

Entered as Second Class Matter Jan. 20, 1926, at the Postoffice at Chicago, Under the Act of Congress of March 3, 1879

COPYRIGHT, 1938, BY THE AMERICAN MEDICAL ASSOCIATION

CONTENTS OF PREVIOUS NUMBER

JULY 1938. NUMBER 1

Preface.

Frontispiece.

Ludvig Hektoen: A Biography and an Appreciation. Morris Fishbein, M.D., Chicago.

Physicians Associated with Ludvig Hektoen from 1897 to 1938.

Bibliography of the Writings of Ludvig Hektoen.

Giant Cells in Cirrhosis. E. Gideon Wells, M.D., Chicago.

Study of Origin of Experimental Gastric Ulcer Induced by Cinchophen. J. P. Simonds, M.D., Chicago.

Physicochemical Status of Protoplasm. Martin H. Fischer, M.D., Pharm.D., Sc.D., and Werner T. Bauer, M.S., Cincinnati.

A New Streptococcus from Dental Caries. Ruth Fosdick, M.D., and Carolyn Hammond, M.A., Chicago.

Isolation of Bacteria from Virus and Phage by a Serial Dilution Method. Edward C. Rosenow, M.D., Rochester, Minn.

Acute Methyl Alcohol Poisoning: A Report of Thirty-Two Instances with Postmortem Examinations. Frank R. Monsé, M.D., Portland, Ore.

Pneumococcemia Anthracia. George F. Dick, M.D., and Alice K. Dick, Ph.D., Chicago.

Preparation and Precipitin Reactions of Some Crystallized Ovalbumins. Arthur G. Cole, Ph.D., Chicago.

Aortic Stenosis, Atresia and Adult Coarctation Associated with a Triloculate Heart. George J. Robertson, M.D., Chicago.

Course of Hematogenous Tuberculous Infection in Guinea Pigs: Pathologic Changes Produced by Massive Intravenous Injections of Virulent and Attenuated Tubercle Bacilli. H. J. Corper, M.D., Denver.

Origin and Pathology of Common Terminal Pneumonia. Virgil H. Moon, M.D., Philadelphia.

Protection by Citrate Against Destruction of Vitamin C by Basicity of Fats of the Diet. Victor Johnson, Ph.D.; Anton J. Carlson, M.D., Ph.D., and Paul Bergstrom, M.D., Chicago.

Use of Polyvalent Antistreptococcus Serum. Eyre. William E. Cary, Ph.D., M.D., Chicago.

Lymphoblastoma of the Rectum. Vernon C. David, M.D., Chicago.

Splenomegaly of the Banti Type: Report of a Case with Postmortem Observations Four Years After Splenectomy. Laurence E. Hines, M.D., and Hermann Fitzgerald, M.D., Chicago.

Reactions to Blood Transfusion: An Attempt at Etiologic Classification; Therapy Based Thereon. Bernard Fantus, M.D.; Lindon Seed, M.D., and Elizabeth Schirmer, M.D., Chicago.

Mixed Tumors of the Lung: So-Called Bronchial or Pulmonary Adenoma. Nathan A. Womack, M.D., and Evarts A. Graham, M.D., St. Louis.

Diverticulitis of the Colon: A Preliminary Report of a Study of Its Potential Role as a Focus of Infection in Systemic Disease. George H. Coleman, M.D., and Joseph A. Capps, M.D., Chicago.

Symmetric Hemorrhagic Necrosis of the Cerebrum Following Primary Thrombosis of the Superior Longitudinal Sinus. Edwin F. Hirsch, M.D., Chicago.

Bacillary Dysentery in Children: A Study of Three Hundred and Fifty-Six Cases from the Children's Division in the Cook County Hospital, Chicago. Maurice L. Flatt, M.D., and Noel G. Shaw, M.D., Chicago.

Pseudomucinous and Apparently Inoperable Papillary Tumor of the Ovaries. Joseph L. Baer, M.D., Chicago.

Hyperemic Inflammation. Frederick Steen, M.D., Chicago.

Fluoroscopy Alone in Diagnosis and Prognosis. W. [illegible]ering, M.D., and R. R. Madison, Ph.D., Stanford University, Calif.

Hemochromatosis: Report of Thirty Cases in Which the Diagnosis Was Made During Life. Hugh R. Butt, M.D., and Russell M. Wilder, M.D., Rochester, Minn.

Synovial Membrane and Fluid in Rheumatoid Arthritis. Edwin P. Jordan, M.D., Chicago.

Gallstone Ileus Syndrome and Related Conditions (Ileus Due to Gallstones, Tuberculomyelitis and Meningomyelocele). Peter Bassoe, M.D., Chicago.

Changes in the Brain from Thyrotoxicosis. I. B. Diamond, M.D., Chicago.

Concentration and Hydration of the Serum Colloids in Disease. Kamil Schulhof, M.D., Chicago.

Clinical Study of Epidemic Poliomyelitis. Lube W. Hess, M.D., Chicago.

Colloid Titration: Methods for the Evaluation of Antigens. Paul G. Heineman, Ph.D., Albany, N. Y.

Colloid (Gelatinoid) Cyst of the Third Ventricle. Eugene F. Traut, M.D., and Eugene C. Pierce, M.D., Chicago.

Nontuberculous Disease of the Upper Lobes of the Lungs Simulating Tuberculosis. B. R. Lovett, M.D., Oak Terrace, Minn.

Goiter and Malignant Growth of the Thyroid in the Dog. David J. Davis, M.D., Chicago.

Mixed Tumors of the Lip. H. E. Eggers, M.D., Omaha.

Cause and Surgical Relief of Sterility in Women. Arthur H. Curtis, M.D., Chicago.

Immunologic Studies on Fractures of Red Blood Cells. [illegible] Delves, Ph.D., Chicago.

Acute Endocarditis Caused by Bacterium Paratyphosum B. Jacob Meyer, M.D., and Katherine M. Howell, M.D., Chicago.

Observations on Guinea Pigs Given Intravenous Injections of Tuberculous Exudates. Russell B. Herrold, M.D., and Carrie I. Wheeler, A.M., Chicago.

Monoblastic Myeloma with Crystalline Protein in the Renal Tubes: Report of a Case. F. D. Gunn, M.D., Winnetka, Ill., and Arthur R. Meeks, M.D., Chicago.

Use of the Fluoroscope in Examination of College Students. Ruth E. Taylor, M.D., Chicago.

Lymphoid Hyperplasia Associated with Thrombocytic Purpura. Laurence E. Cooley, M.D., Dubuque, Iowa.

Sensitiveness to Insulin and Optimal Diets in Diabetes. Walter H. Nadler, M.D., Chicago.

Clinical and Statistical Study of Sore Throat in Young Adults. Paul S. Rhoads, M.D., Evanston, Ill., and M. L. Afremow, M.D., Chicago.

Atherosclerosis of the Coronary Arteries. Daniel J. Glomset, M.D., Des Moines, Iowa.

Epithelial Cyst of the Heart. Israel Davidsohn, M.D., Chicago.

Sarcomatous Degeneration of Transplantable Mammary Adenofibroma of the White Rat. Ludwig A. Emge, M.D., San Francisco.

Crystalline Bence Jones Proteins. Arthur R. Meeks, M.D.; Lindon Seed, M.D., and William E. Welker, Ph.D., Chicago.

ARCHIVES OF PATHOLOGY

VOLUME 26 AUGUST 1938 NUMBER 2

COPYRIGHT, 1938, BY THE AMERICAN MEDICAL ASSOCIATION

CHOLECYSTITIS AND HYPERTROPHY OF THE MUSCULARIS OF THE GALLBLADDER

KNOWLES B. LAWRENCE, M.D.

AND

SHIELDS WARREN, M.D.

BOSTON

The muscular tunic of the human gallbladder, like other visceral smooth muscle, is capable of hypertrophy. In this study measurements of the thickness of the muscular layer in a series of 100 surgically removed gallbladders demonstrate the frequent association of hypertrophy with chronic cholecystitis and cholelithiasis.

MATERIAL

The material used in the study included a series of gallbladders removed surgically and other gallbladders obtained post mortem, the latter as normal controls. The surgical series consisted of 100 consecutive specimens obtained at the New England Deaconess Hospital and the New England Baptist Hospital between September 1936 and May 1937. Only a few gallbladders in the series were excluded, those in which the muscle could not be measured satisfactorily, because of extreme inflammatory changes or other factors. The single gallbladder showing no microscopic evidence of cholecystitis was excluded also. The material obtained post mortem consisted of 50 gallbladders from autopsies made at these hospitals during the same period. This group did not represent a consecutive series but included only gallbladders that were normal microscopically and without stones.

Sections were taken usually from the midportion of the peritonealized wall of the fundus, occasionally from other parts. The microscopic slides of the surgical series were prepared by a forty-eight hour method of alcohol-formaldehyde fixation and preparation of pyroxylin sections. For the postmortem material a two week method of fixation in Zenker's solution and preparation of paraffin sections was used.

To determine what discrepancies in the thickness of the muscle might arise from the use of the different technic, a series of 30 gallbladders, including 10 normal and 20 diseased specimens, were prepared by both technics. The averages of the measurements of the thickness of the muscle in this series yielded values of 0.35 mm. for paraffin and 0.40 mm. for pyroxylin sections, a ratio of 7:8. The greatest difference existed in the normal group, which gave 0.14 mm. for the paraffin and 0.21 mm. for the paroxylin sections, or a ratio of 2:3.

From the Laboratory of Pathology, New England Deaconess Hospital.

Almost all the surgically removed gallbladders were received at the laboratory opened, empty of bile, wrapped in gauze wet with physiologic solution of sodium chloride and within a few hours after operation. In consequence their preservation was good, but some possibility of postmortem contraction with apparent thickening of the muscle was not eliminated. It was felt that adequate compensation for this possibility could be achieved by the use of a factor based on the relative areas of the surgically removed and the normal gallbladder. The ratio of the average area of the surgically removed organs to the average area of the gallbladders obtained post mortem when these were opened and spread out was, in square centimeters, 41 : 55.

The microscopic slide sections in each case represented cuts approximately perpendicular to the lumen of the gallbladder. With an ocular micrometer, the thickness of the muscular coat was measured in hundredths of a millimeter. Three or more measurements were taken at regular intervals along the muscular layer, approximate allowances being made for separation of muscle bundles, due to fibrosis, inflammatory reaction and shrinkage during fixation. These various measurements were averaged to give the thickness of the muscularis of each specimen.

RESULTS

The table summarizes part of the data obtained. The average thickness of the muscular layer as determined in the normal gallbladders

Data from the Study of Normal and Chronically Inflamed Gallbladders

	Normal Gallbladders Observed Post Mortem (Total Number 50)	Surgically Removed Gallbladders with Chronic Cholecystitis (Total Number 100)
Average area of wall (measured fresh), sq. cm...	55	41
Average thickness of musculature layer, mm....	0.16	0.54
Average thickness of muscle corrected for postmortem contraction and shrinkage in fixation, mm.	..	0.35
Number showing Rokitansky-Aschoff sinuses....	3	60
Number with calculi.....	0	90

obtained post mortem was 0.16 mm. and as determined in the surgically removed gallbladders, all of which showed some degree of cholecystitis, was 0.54 mm. The average areas of these same two groups of gallbladders were, however, 55 and 41 sq. cm., respectively. Multiplying the factor 41/55 (the relative wall areas) by 7/8 (the shrinkage factor) by 0.54 (the average thickness of muscle in the surgically removed gallbladders) gave an adjusted value of 0.35 mm. for the muscle in the surgically removed gallbladders. After making adequate adjustments for possible artefacts, as outlined, the muscular coats in the diseased gallbladders were found to be more than twice as thick as those in normal gallbladders (0.35 and 0.16 mm., respectively). These figures, however, do not do justice to the true state of affairs since 74 of the 100 surgically removed gallbladders revealed well defined hypertrophy of the muscle microscopically: that is, a thickness greater than the upper extreme of the normal gallbladder (0.32 mm.). Twenty-eight of these showed marked hypertrophy, measuring 0.7 mm. or more in thickness.

Four gallbladders had a thickness of the muscle ranging from 1 to 1.5 mm. Only a fraction of this increase could be attributed to contraction, since the areas of these gallbladders averaged 42 sq. cm. In 26 of the 100 gallbladders the musculature was either normal or not conclusively hypertrophied microscopically, the thinnest measuring 0.12 mm. The extremes for the normal postmortem series were 0.7 and 0.32 mm.

A study was made of the mucosal diverticula which are found frequently in thickened gallbladders and have been called Rokitansky-Aschoff sinuses.[1] Herniation of the mucosal pockets through at least one muscle layer was the criterion employed. In a number of cases the pockets penetrated a greatly hypertrophied muscularis, even extending short distances into the serosal stroma (fig. 2). There were well

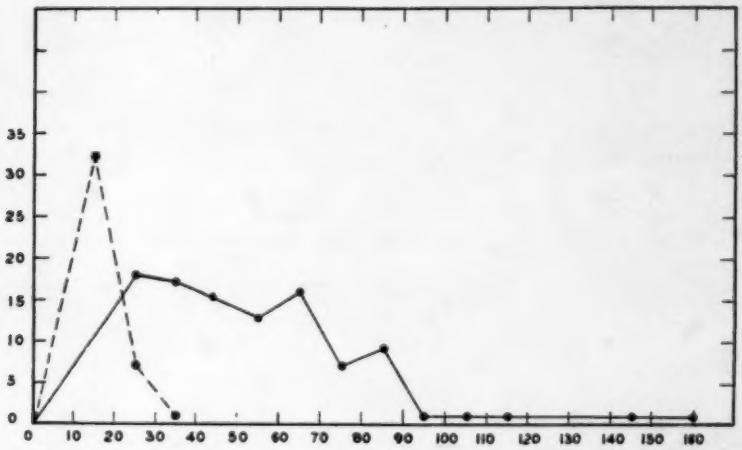


Fig. 1.—The thickness of the musculature in normal and in chronically inflamed gallbladders. The numerals at the left represent the numbers of specimens; those at the bottom of the chart, the thickness of the musculature in hundredths of a millimeter. The broken line represents normal gallbladders, observed post mortem; the solid line, surgically removed gallbladders with chronic cholecystitis.

formed Rokitansky-Aschoff sinuses in 60 of the 100 surgically removed gallbladders but in only 3 of the 50 normal gallbladders. Figures 2 and 3 illustrate slight and marked hypertrophy of the musculature of the gallbladder in cases of chronic cholecystitis. Several Rokitansky-Aschoff sinuses are shown.

The presence of calculi was noted also, although no attempt was made to classify the stones as to type. As stated already, none of the gallbladders obtained post mortem contained stones. In 90 of the 100 cases in which a surgically removed organ was examined, there were

1. Halpert, B.: Bull. Johns Hopkins Hosp. **41**:77, 1927.

one or more calculi in the extrahepatic biliary system. The great majority of the stones were within the gallbladders and were of the multiple mixed type.

COMMENT

The observation of muscular hypertrophy in the human gallbladder showing chronic cholecystitis has been recorded by a number of writers. Nuboer,[2] Mackey[3] and Graham and Mackey[4] described or mentioned the hypertrophy but were unable to correlate it significantly with the

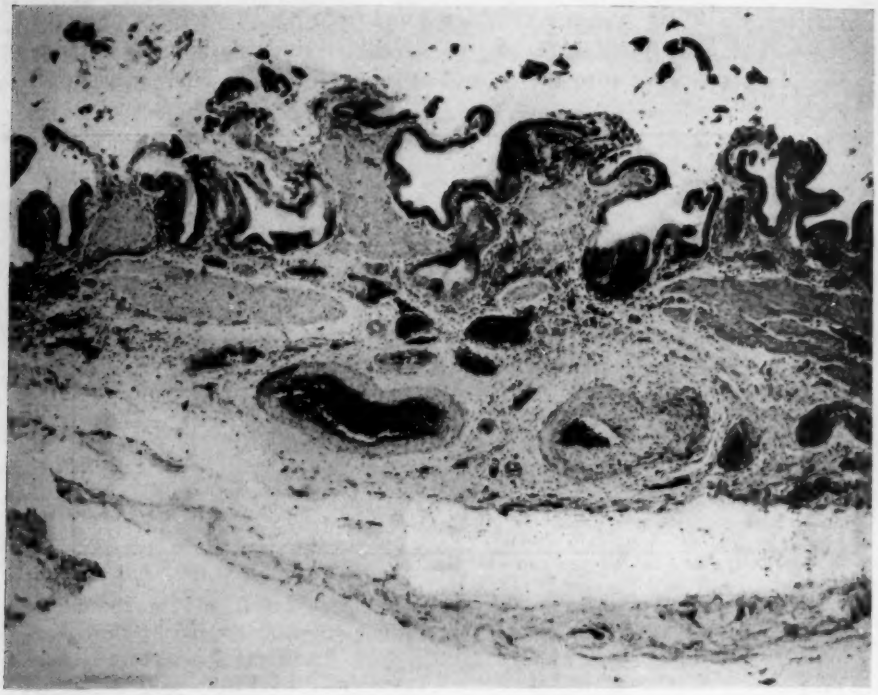


Fig. 2.—Section from the fundus of a chronically inflamed gallbladder showing slight muscular hypertrophy (0.4 mm. thick) with shallow but numerous Rokitansky-Aschoff sinuses; × 48.

disease process. Andrews[5] found muscular hypertrophy of from slight to 2 plus grade in only 16 of 54 gallbladders surgically removed and felt that it was seldom important. Burden[6] thought that partial obstruc-

2. Nuboer, J. F.: Frankfurt. Ztschr. f. Path. **41**:454, 1931.
3. Mackey, W. A.: Glasgow M. J. **115**:225, 1931.
4. Graham, E. A., and Mackey, W. A.: J. A. M. A. **103**:1497, 1934.
5. Andrews, E.: Arch. Surg. **31**:767, 1935.
6. Burden, V. G.: Ann. Surg. **85**:239, 1927.

tion to the outflow of bile, possibly at the neck of the gallbladder, accounted for the muscular hypertrophy observed by him. One of us (Lawrence), with Halpert, in a paper [7] correlating pathologic changes in the gallbladder with types of gallstones, noted the frequent occurrence of muscular hypertrophy in chronic cholecystitis. Halpert [1] described

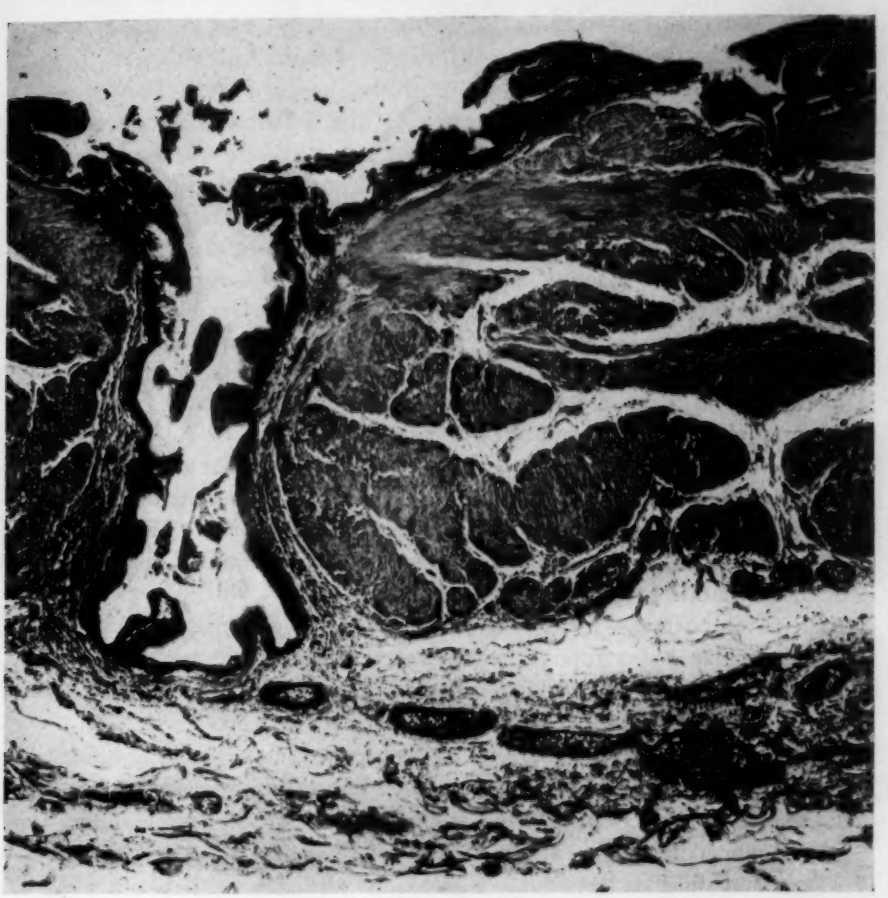


Fig. 3.—Section from the fundus of a chronically inflamed gallbladder showing marked muscular hypertrophy (1.2 mm. thick) and a Rokitansky-Aschoff sinus extending through the muscularis into the subserosa; × 48.

hypertrophy in association with the presence of Rokitansky-Aschoff sinuses and presented several good photomicrographs in illustration of this finding.

7. Halpert, B., and Lawrence, K. B.: Surg., Gynec. & Obst. **62**:43, 1936.

No figures for the percental occurrence of Rokitansky-Aschoff sinuses were given by the writers who mentioned this phenomenon. The hypothesis concerning the mode of formation of such sinuses given by Halpert [1] is compatible with the high percentage found by us in the chronically inflamed gallbladders: " . . . in the process of formation of deeper outpouchings of the gallbladder mucosa toward the external layers, extreme contractions following prolonged and repeated overdistentions of the viscus are most likely to play the leading part."

The high frequency of gallstones in instances of chronic cholecystitis has been pointed out in the literature.[8] The findings in this study are merely confirmatory.

In addition to the increase in the thickness of the muscularis as measured in this investigation, the hypertrophied muscle in the chronically diseased gallbladders presented some interesting features. Individual muscle fibers were sometimes much thicker and longer than in the normal controls. The nuclei of the muscle cells showed considerable variation in size, being in some cases from four to six times normal. The interwoven fascicular pattern apparent in control normal gallbladders was exaggerated in gallbladders showing marked muscular hypertrophy, despite dilatation in some cases.

Boyden [9] pointed out that in the cat's gallbladder contracted or collapsed after a meal of fat the thickness of the muscularis may be increased ten times over that of the dilated gallbladder. This increase is accompanied, however, by a nineteen-fold thickening of the mucosa. In none of our gallbladders did the mucosa show a thickening of more than twice normal. In fact, as the chronic cholecystitis increased and the muscular hypertrophy became more marked, the usual papillary infoldings of the mucosa tended to flatten out, with loss of the normal rugal pattern.

This observed hypertrophy of the muscularis in chronically inflamed gallbladders may well be due to increased intrabiliary pressure from (*a*) mechanical obstruction, as from stone, stricture or cholangitis, and (*b*) physiologic defects, as failure of absorption of vesical contents, reflex spasm and the like.

That several such factors are involved seems likely although difficult to prove. Clinical studies on part of the material yielded clues to the etiologic explanation of the muscular hypertrophy in certain cases but were inconclusive statistically.

---

8. Baumgartner, C. J.: Surg., Gynec. & Obst. **49**:780, 1929. Denton, J.: Arch. Surg. **14**:1, 1927. Crump, C.: Surg., Gynec. & Obst. **53**:447, 1931. Andrews.[5] Halpert and Lawrence.[7]

9. Boyden, E. A.: Anat. Rec. **30**:333, 1925.

SUMMARY

Measurements were made of the thickness of the muscularis in a series of 100 surgically removed gallbladders, all of which showed some degree of chronic cholecystitis. A control series of 50 normal gallbladders from autopsies was employed. Adequate allowances were made for apparent variations in the muscle thickness due to relative size and physiologic state of the gallbladder, to postmortem shrinkage of tissues and to the use of different technics of preparation. The average thickness of the muscularis in the chronically inflamed gallbladders was found to be more than twice that in the normal gallbladders. Microscopically, this increase appeared due to definite hypertrophy of the smooth muscle fibers.

No satisfactory etiologic explanation could be postulated for the muscular hypertrophy as a result of this study.

EXPERIMENTAL HYPERCHOLESTEREMIA IN DOGS

WARREN C. CORWIN, M.D.
Fellow in Experimental Surgery, the Mayo Foundation
ROCHESTER, MINN.

Anitschkow and Chalatow,[1] working independently of others, and Wacker and Hueck,[2] also working independently of others, reported almost simultaneously in 1913 that pure cholesterol administered by mouth over a considerable period could produce lesions in the intima of the aorta of the rabbit very similar to those of arteriosclerosis of man. The work of these investigators was referred to by Duff [3] in his excellent review of experimental cholesterol arteriosclerosis. The observation has been confirmed, moreover, by numerous investigators. Bailey [4] in 1915, and several other workers since, have reported the production of arteriosclerosis in guinea pigs by the feeding of cholesterol, but there has been no convincing demonstration of a similar effect in any other species. It is true that lesions have been described as occurring in the arteries of pigeons, chickens, parrots, mice, rats and goats following feeding of cholesterol, but because of the inconstancy of their occurrence they must be regarded with a strong suspicion that they were spontaneous in origin. Up to the present time success has not attended any attempt to produce the condition known as cholesterol arteriosclerosis in cats, dogs, foxes and monkeys.

Duff [3] expressed the opinion that hypercholesteremia of some degree is an essential factor in the development of experimental cholesterol arteriosclerosis of rabbits. If this is true, the failure to produce arteriosclerosis in the dog by feeding a diet rich in cholesterol may be assumed to be owing at least partly to the inability of investigators to produce hypercholesteremia consistently in this animal. Grigaut and L'Huillier [5] found moderate hypercholesteremia in 2 dogs each of which received five daily feedings of cholesterol dissolved in melted butter and mixed with meat. By daily feeding of cholesterol and bile, Anitschkow [6] was able to raise to about twice its normal value the cholesterol content of

1. Anitschkow, N., and Chalatow, S.: Centralbl. f. allg. Path. u. path. Anat. **24**:1, 1913.
2. Wacker, L., and Hueck, W.: (*a*) München. med. Wchnschr. **60**:2097, 1913; (*b*) Arch. f. exper. Path. u. Pharmakol. **73**:416, 1913.
3. Duff, G. L.: Arch. Path. **20**:81 and 259, 1935.
4. Bailey, C. H.: Proc. Soc. Exper. Biol. & Med. **13**:60, 1915-1916.
5. Grigaut, A., and L'Huillier, A.: Compt. rend. Soc. de biol. **73**:304, 1912.
6. Anitschkow, N.: Verhandl. d. deutsch. path. Gesellsch. **20**:149, 1925.

the blood of a dog which had been subjected to partial ligation of the common bile duct, ovariectomy and splenectomy. Weltmann and Biach[7] found no increase in the concentration of cholesterol in the blood of a puppy following feeding of cholesterol for sixteen days, and Ssokoloff[8] likewise found none in 10 dogs after they had been fed cholesterol for seven days. Muñoz[9] reported that the concentration of cholesterol in the blood of the dog is higher when the diet is of meat than when it is high in fat or carbohydrate.

FIRST SERIES OF EXPERIMENTS

Hueck and Wacker,[10] Versé[11] and Page and Bernhard[12] showed that hypercholesteremia is invariably associated with sufficient increase in neutral fats and in other lipoids of the blood to cause marked lipemia. At the outset of the experimental work to be reported here, therefore, it was deemed desirable to determine what effect on the cholesterol content of the blood might be obtained by the feeding of diets high in fat without added cholesterol, bile salts and lecithin as well as with these substances added in various combinations.

Accordingly, 9 adult dogs, ranging from 12 to 18 Kg. in weight, were chosen, and in preliminary studies normal values for blood cholesterol were set up for dogs fed on the two types of diet most commonly used in this laboratory, namely, the standard diet and the kennel diet. The former diet consists of a mixture of the following foods in the percentages given: horse meat, 38; cracker meal, 35; canned tomatoes, 15; lard, 8, and bone ash, 4. A weighed quantity was given daily. The latter diet consists of dog biscuits, alternating with a mash made of ground horse meat, corn meal, oat meal and bone ash. Determinations of cholesterol were made by the Lieberman-Burchard method as used by Bloor and his associates.[13] The values for the blood cholesterol of 4 dogs maintained on the standard diet for six weeks ranged from 185 to 285 mg., with an average of 225 mg., per hundred cubic centimeters. The concentration of the blood cholesterol of 5 dogs maintained on the kennel diet for the same length of time ranged from 155 to 240 mg., with an average of 195 mg., per hundred cubic centimeters. The values for the blood cholesterol of the same dog varied somewhat from hour to hour, but a characteristic diurnal curve was not obtained. As a rule the highest value was reached from three to six hours after the daily meal, particularly if the dogs were maintained on the standard diet, but this was not observed invariably.

7. Weltmann, O., and Biach, P.: Ztschr. f. exper. Path. u. Therap. **14**:367, 1913.

8. Ssokoloff, N. A.: Virchows Arch. f. path. Anat. **245**:203, 1923.

9. Muñoz, J. M.: Rev. Soc. argent. de biol. **8**:595, 1932.

10. Hueck, W., and Wacker, L.: Biochem. Ztschr. **100**:84, 1919-1920.

11. Versé, M.: Beitr. z. path. Anat. u. z. allg. Path. **63**:789, 1917.

12. Page, I. H., and Bernhard, W. G.: Arch. Path. **19**:530, 1935.

13. Bloor, W. R.; Pelkan, K. F., and Allen, D. M.: J. Biol. Chem. **52**:191, 1922.

RESULTS IN FIRST SERIES

After the control values had been determined, the animals were placed on the following dietary regimens for eight weeks, with results as described:

Group 1.—This group was made up of 3 dogs which received high fat diet A daily. This diet consisted of 4 Gm. per kilogram of body weight of the standard diet described and 10 Gm. per kilogram of body weight of lard.

This diet was effective in producing moderate hypercholesteremia in 2 of the 3 dogs of this group. Several readings of the cholesterol content of the blood of dog A exceeded 400 mg. per hundred cubic centimeters; the highest reading was 482 mg. per hundred cubic centimeters, while the value for the blood cholesterol of dog B ranged consistently around 350 mg. during the last week of the feeding. Dog C at no time exceeded the upper limit of normal cholesteremia as determined after feeding the standard diet in preliminary experiments.

Group 2.—Dog D was maintained on high fat diet A, and 3 Gm. of solid cholesterol was given by capsule daily. Dog E received high fat diet A and 3 Gm. of sodium cholate daily, while dog F was given high fat diet A, cholesterol and sodium cholate.

The concentration of the blood cholesterol of all of the dogs of this group became moderately augmented. The augmentation, however, did not exceed that which, according to results obtained in group 1, might have been expected from feeding the high fat diet alone. During the last week of the experiment the values for the blood cholesterol of all 3 dogs ranged from 300 to 400 mg. per hundred cubic centimeters.

Group 3.—These 3 dogs each received from 10 to 15 Gm. daily of lecithin of adrenal origin, prepared and furnished by Dr. E. C. Kendall. In addition, dog G received high fat diet A and cholesterol, dog H received high fat diet A and sodium cholate, and dog I received high fat diet A, cholesterol and sodium cholate.

In the 3 dogs of this group hypercholesteremia of considerable proportions developed. The blood cholesterol of each rose to more than 600 mg. per hundred cubic centimeters on more than one occasion during the last two weeks of the experiment. In dog G, which received both cholesterol and lecithin, the value for blood cholesterol on three separate occasions exceeded 800 mg. per hundred cubic centimeters; once the value was 940 mg.

COMMENT ON FIRST SERIES

It is probably safe to conclude from this first series of experiments that diets of which the fat content is very high may produce moderate hypercholesteremia, and that lecithin administered in conjunction with a high fat diet will produce hypercholesteremia of considerable proportions. It should be noted that the fat of the diet employed, lard, is a pure fat and that therefore the effect obtained cannot be attributed to vitamins, phospholipins or other lipoid substances. In all probability neither cholesterol nor sodium cholate has any effect on the blood cholesterol of the dog, at least when administered by mouth in the solid state.

There is no doubt that the feeding of large amounts of fat would be an excellent method of producing hypercholesteremia in dogs if it

were not for the fact that most dogs have for such diets a marked intolerance, expressed by discouragingly frequent emesis. Perhaps the relation between ingestion of fat with actual retention and the development of hypercholesteremia may be brought out best by stating that dog C (group 1), which retained practically no food during the last two weeks of the experiment, showed no increase in the concentration of blood cholesterol, whereas dog G (group 3), which vomited not more than two or three times throughout the entire period of eight weeks, presented the most marked hypercholesteremia seen to date in this laboratory.

SECOND SERIES OF EXPERIMENTS

Because the lecithin employed in the previous experiments apparently was efficacious in raising the concentration of the blood cholesterol of dogs, it seemed desirable to investigate its effect still further.

To avoid confusing dogs of the first series of experiments with those of the second series, dogs of the latter series were designated by arabic numerals. Seven dogs were given the standard diet. One dog (dog 8) received the kennel diet for a control period of two weeks. On the seventh and fourteenth days, determinations of cholesterol were made on the blood withdrawn following a fast of twenty-four hours and on blood withdrawn five hours after feeding. The average of the four readings thus obtained was taken as a standard control value of the blood cholesterol for the particular dog in question. The dogs were then given the special diets and medications to be enumerated, and the values for the blood cholesterol were obtained following a fast of twenty-four hours and again five hours after feeding. The values were determined at weekly intervals for an experimental period of six weeks. The administration of the special diets and the medications were then discontinued, and the values for the blood cholesterol were determined at weekly intervals for a recovery period of four weeks.

During the experimental period the 8 dogs were maintained as follows: Dogs 1 and 2 received high fat diet B (fig. 1). This diet consisted of 10 Gm. of the standard diet per kilogram of body weight plus 5 Gm. of lard per kilogram of body weight. This was tolerated much better than high fat diet A because it had a much lower content of fat. High fat diet B contained 33 per cent lard by weight, as against 71.5 per cent in high fat diet A. Dogs 3 and 4 were given high fat diet B plus lecithin (fig. 2). To dogs 5 and 6 the standard diet plus cholesterol and lecithin was administered (fig. 3). Dog 7 was given the standard diet plus lecithin (fig. 4). Dog 8 received the kennel diet plus lecithin (fig. 4).

RESULTS IN SECOND SERIES

From a brief perusal of the accompanying figures it may be concluded that a diet high in fat may or may not effect a rise in the concentration of cholesterol in the blood of the dog (fig. 1). This is in agreement with the findings in the first series of experiments. Lecithin when added to a diet high in fat will produce marked hypercholesteremia (fig. 2). In the opinion of others who observed this work and in my opinion the experiments recorded here were not carried out for a sufficient length of time to determine the true proportions to which

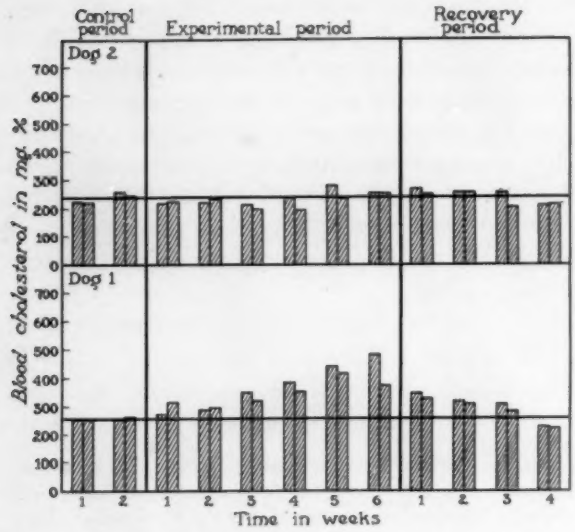


Fig. 1.—Effect of feeding high fat diet B. In this figure and in figures 2, 3 and 4 the first block of each pair represents the cholesterol content of the blood after a fast of twenty-four hours; the second block, the cholesterol content five hours after feeding. The horizontal line represents the average blood cholesterol during the control period.

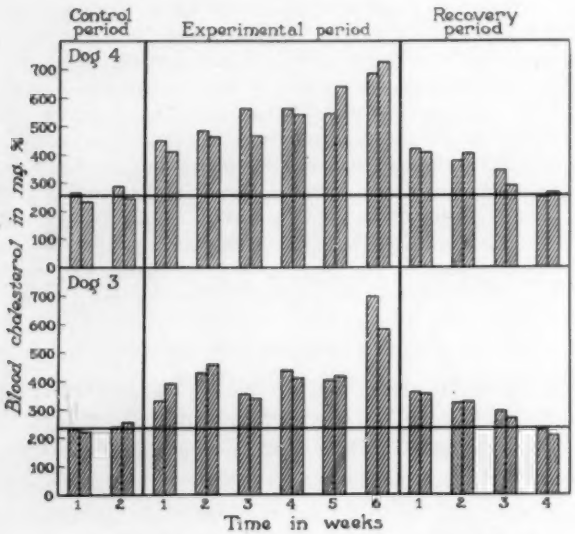


Fig. 2.—Effect of feeding high fat diet B and lecithin.

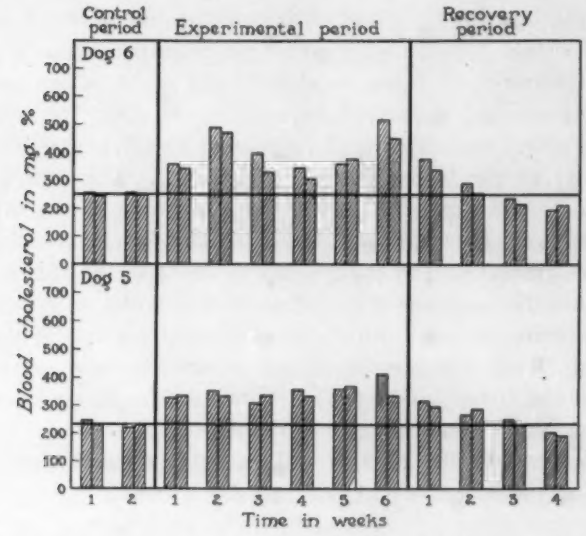


Fig. 3.—Effect of feeding the standard diet, lecithin and cholesterol.

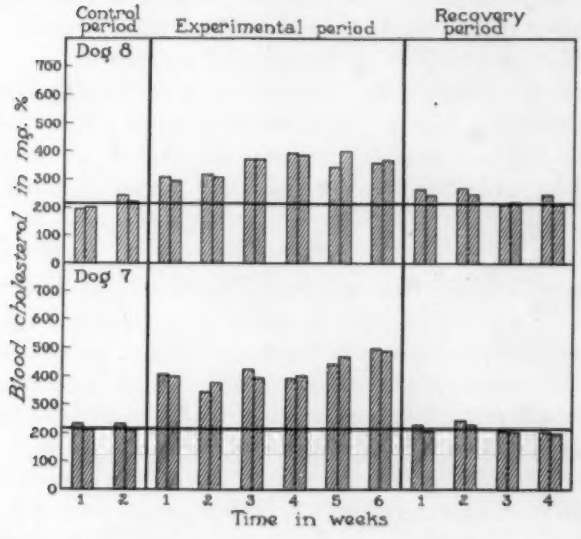


Fig. 4.—Effect of feeding the standard diet and lecithin (dog 7) and the kennel diet and lecithin (dog 8).

such hypercholesteremia may attain. At the end of the experimental period the values for the blood cholesterol of dogs 3 and 4 were apparently still rising. Obviously, more extended experiments are desirable.

As is indicated in figure 4, a diet high in fat is not essential for the development of hypercholesteremia in the dog, for when lecithin alone was added to either the standard diet or the kennel diet elevation of the level of the blood cholesterol occurred. The addition of solid cholesterol to this regimen did not materially alter the results (fig. 3).

Wacker and Hueck,[2a] and Versé [11] reported that in hypercholesteremia the greater part of the increase is owing to the rise in cholesterol esters, while the augmentation of free cholesterol is not so striking. In my experiments, determinations of cholesterol esters, made by the method of Bloor and Knudson [14] on random samples of blood, indicated that the hypercholesteremia is owing to a rise in concentration of both the cholesterol esters and the free cholesterol. The value for free cholesterol in the majority of instances ranged from 30 to 40 per cent of the value for total serum cholesterol.

COMMENT ON THE SECOND SERIES

If the development of arteriosclerosis in the dog is dependent on the development of hypercholesteremia, as is the case with the rabbit, the studies reported here indicate that perhaps the dog may yet be employed in the study of this disease. At present I am maintaining a small group of dogs on long time feeding with lecithin. Studies of cholesteremia resulting from feeding of lecithin derived from eggs also are being conducted in this laboratory. Since other investigators have demonstrated lipemia in association with hypercholesteremia,[15] the neutral fats and phospholipoids of the blood were followed in my second series of experiments. These are to be reported in a later paper.[16]

SUMMARY

The data obtained from the experiments indicate that a diet high in fat may or may not produce moderate hypercholesteremia in the dog. Lecithin of adrenal origin, when given by mouth daily, will constantly raise the concentration of cholesterol in the blood. Lecithin plus a diet high in fat will effect marked elevation of the level of the blood cholesterol. This elevation is owing to increased concentration both of free cholesterol and of cholesterol esters. Cholesterol and sodium cholate have no effect when administered by capsule in the solid state.

---

14. Bloor, W. R., and Knudson, A.: J. Biol. Chem. **27**:107, 1916.

15. Hueck and Wacker.[10] Versé.[11] Page and Bernhard.[12]

16. Flock, E. V.; Corwin, W. C., and Bollman, J. L.: Sustained Hyperlipemia of Dietary Origin in the Dog, Am. J. Physiol., to be published.

DESTRUCTION OF THE GANGLION CELL IN THE INFANT BRAIN

C. R. TUTHILL, M.D.
STATEN ISLAND, N. Y.

Karyorrhexis of the nuclei of ganglion cells was considered by Spielmeyer [1] to be a nuclear disturbance accompanying severe change of ganglion cells in cerebral inflammation. The nuclear protoplasm is broken up into many fine granules, which are seen either within or without the nuclear periphery; the nucleolus remains intact, and the cytoplasm is swollen and tygrolytic. The nuclear fragments finally fade, and the cell disappears. Low [2] described karyorrhexis of the nuclei of ganglion cells in "toxic encephalitis" as fine granules formed around the periphery of the nucleus. The protoplasm within the nucleus is homogeneous, the nucleolus is unchanged, while the cytoplasm is pale and watery. The end of this cell is indicated by hypochromatic nuclear fragments, also. Karyorrhexis of the astrocytes,[1] on the other hand, is recognized as a fairly uniform change in which small divisions of the nucleus are formed in a shrunken cytoplasm, which is either palely or distinctly stained. In a study of infant brains a disintegration of the ganglion cells was found which resembled the karyorrhexis in astrocytes rather than that which has previously been described in ganglion cells. This has been designated as karyorrhexis of ganglion cells, since the cytoplasmic changes are apparently dependent on the break-up of the nuclei.

MATERIAL

The brains of thirty-four children were examined, ranging in age from 7 days to 2 years. The sections were the usual ones of the cortex, basal ganglions and cerebellum, stained with thionine blue.

OBSERVATIONS

The changes in the ganglion cells in this condition are as follows: The nucleus shows a partition into three or more round, dark blue staining masses, somewhat larger than the nucleolus, and the cytoplasm is shrunken. The periphery of the nucleus is lost, as is the nucleolus, while the particles are scattered over a greater area of the cytoplasm

From the Richmond Memorial Hospital.

1. Spielmeyer, W.: Histopathologie des Nervensystems, Berlin, Julius Springer, 1922, vol. 1.

2. Low, A. A.: Arch. Neurol. & Psychiat. **23**:696, 1930.

than is occupied by a nucleus of the usual size and give the cell a leopard-like spotted appearance. The decreased cytoplasm is more distinctly stained than that in the apparently normal cell and is sometimes of such deep color as to interfere with the clear differentiation of the nuclear divisions. Small dark protoplasmic fragments may also

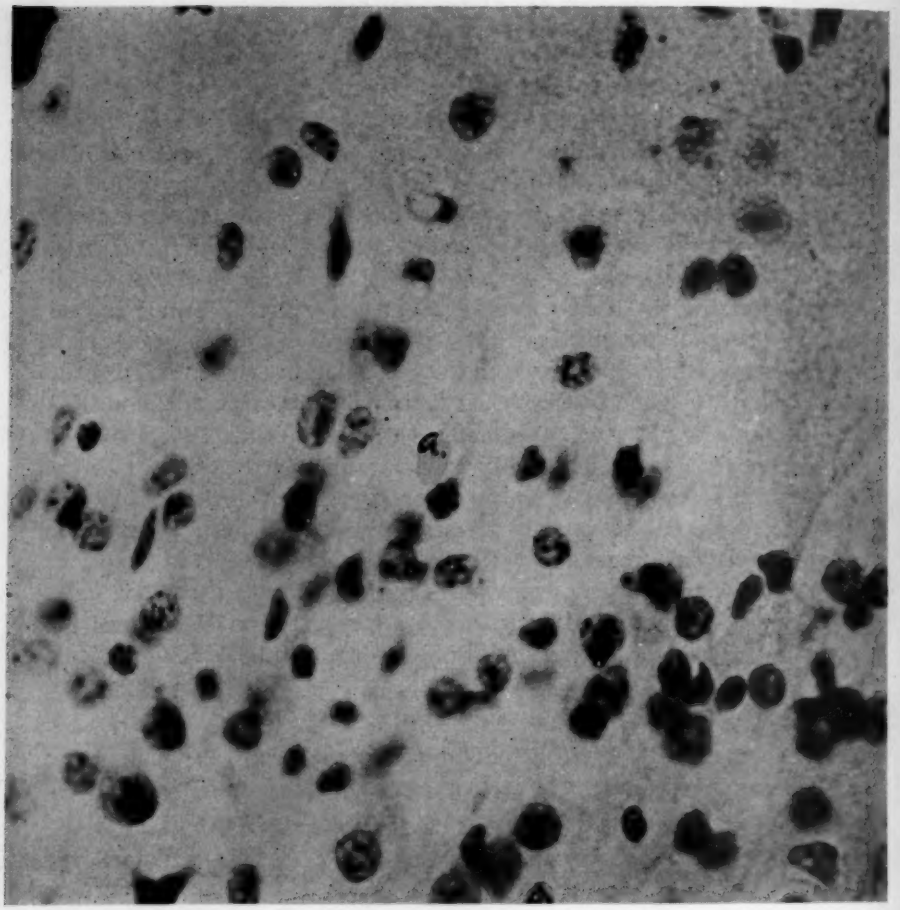


Fig. 1.—The letter *a* indicates a ganglion cell in the second cortical layer with little, although distinctly stained, cytoplasm and nodular dispersion of the nuclear protoplasm.

be found bordering the periphery of the cell. Cells in which the nuclei are broken up in scanty, although sharply and clearly stained, cytoplasm are found in figures 1 and 2. The changes in the cytoplasm are also evident in cell *a* of figure 3, but those of the nucleus are out of focus.

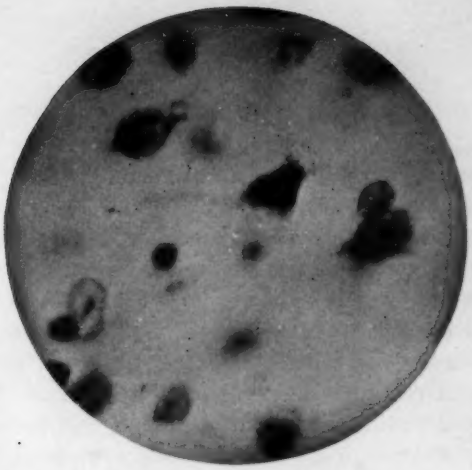


Fig. 2.—A fairly large-sized cortical ganglion cell with numerous nuclear particles in a shrunken, clearly stained cytoplasm.

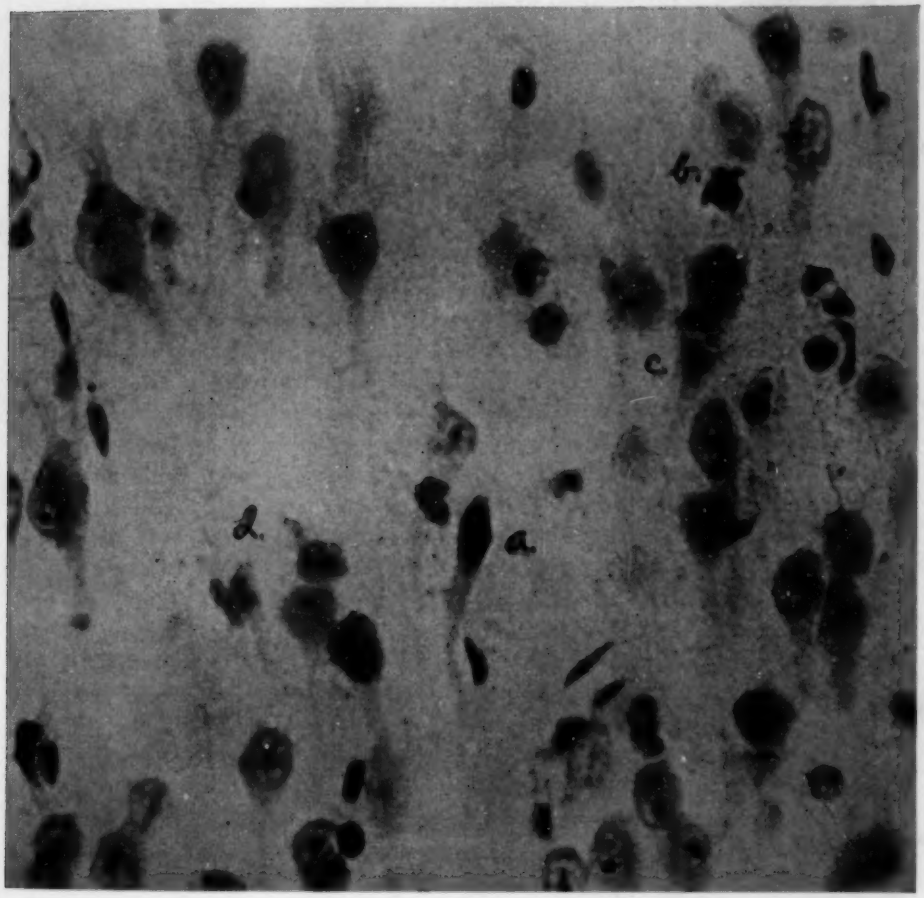


Fig. 3.—Cornu ammonis. Cell *a* is a cell shrunken to one much smaller than the usual size found in this area. The cytoplasm is darker, and the nuclear particles, unfocused, are represented as a deep staining mass. Cells *b* and *c* have similar nuclear particles, which are more distinct against the pale, thin, shrunken cytoplasm. Near a hypertrophic microglia cell is cell *d*, in which both the nuclear fragments and the cytoplasm are faded and shrunken.

With the continuation of the cellular disintegration, the cytoplasm becomes thin and stains feebly, as in cells *b* and *c* of figure 3, although the nuclear particles are apparently untouched. All parts may decrease in size without loss of staining ability and undergo neuronophagia. Other cells fade out in a fashion similar to that observed in cell *d* of figure 3, which has all but disappeared, with its scarcely perceptible nuclear partitions and depleted cytoplasm, while the proximity of an enlarged microglia cell foretells the end of the involved ganglion cell. It is possible that fading is an early change, but as poorly staining nuclear fragments are found only with pale cytoplasm, it seems more probable that the cytoplasm first deepens in color. All of the described cells are best seen when the time post mortem is sufficiently short to permit some preservation of the cytoplasm. When the time post mortem is prolonged, it is not unusual to find the cytoplasm so severely autolyzed that the nuclear spherules are left naked. Or if the cytoplasm is still present, it is somewhat oval in shape, staining a deep blue, and the nuclear particles may be one or two fairly large masses.

Most of the karyorrhexic ganglion cells are found in the cortex. Occasionally such cells are observed in the cornu ammonis and in the subcortical white substance. They are rare in the basal ganglions and are never observed here after the first year. In the cerebellum, they occur among the small ganglion cells of the molecular layer during the first month, while nuclear particles may also be observed between the Purkinje cells. It is difficult to know which size cortical cell is most affected, since the cell is small whenever the change is sufficiently marked for recognition, even among the giant cells of the fifth layer. Rows or groups of cells are not disintegrated, only single ones between apparently normal cells. Although no predilection for special areas in the cortex occurs, the number of affected cells varies in different parts from brain to brain. In the older brains more of the karyorrhexic ganglion cells are found in the second layer, while in the younger ones they occur about equally in all layers but are prominent in the rim of the second layer and in the sparsely cellular lower sixth. The greatest number are observed during the first six months, when from 2 to 6 cells may be found in fairly close fields. Most of the brains of this age, however, show 1 affected cell in about 20 fields. After six months this is reduced with variations to a single cell in many gyri.

Since post mortem changes are present in the brains of all the infants aged up to 2 months, postmortem dissolution has probably destroyed some of the karyorrhexic ganglion cells as it has the others, yet in four cases karyorrhexis of the ganglion cells is at its maximum. The associated conditions in these cases are: cerebral hemorrhage, in an infant aged 10 days; intestinal toxemia, in one aged 19 days; septicemia, in one

aged 17 days, and cellulitis, in one aged 7 weeks. A few disintegrating cells are also observed in the thalamus of the last infant. Other conditions in infants of this age, in which karyorrhexis of the ganglion cells was less marked are: duodenal hematoma, in one aged 7 days; hydrocephalus, in two aged 7 and 15 days; erysipelas, in one aged 14 days; congenital defect of the heart, in one aged 24 days; intestinal toxemia, in one aged 5 weeks; and in four infants aged 7 weeks, adrenal abscess, tumor of the pharynx, bronchopneumonia and erysipelas, respectively. In this group, in the brains of the infants with the first and the last-named condition the nucleus caudatus and thalamus disclose an occasional karyorrhexic ganglion cell.

Among the brains of infants from 2 to 6 months of age are three with fewer cells lost by postmortem dissolution, as not only was the postmortem time short but the increased age prevented the swift descent of autolysis. In two of these brains and in another with a longer postmortem time, karyorrhexis of the ganglion cells is as marked as in the more affected brains of the younger ones. The first of the two is the brain of an infant aged 2 months, with amyotonia congenita; the second is that of an infant aged 3½ months, with rash and convulsions, while the other is that of an infant aged 6 months, with septicemia. In the rest of the group of this age, karyorrhexis of the ganglion cells is less frequent and variable. The associated conditions are: pulmonary atresia, in an infant aged 3 months; intestinal toxemia, in one aged 4 months (this brain is the third mentioned as examined a short time post mortem); acute meningitis, in two aged 5 and 6 months, and tuberculous meningitis, in an infant aged 6 months, in whose brain karyorrhexic ganglion cells are also present on the edge of an area of softening in the thalamus.

Only the minimal number of karyorrhexic ganglion cells occur in the older brains regardless of whether the time post mortem was sufficiently short for fair preservation of the cells or not. The diagnoses are: whooping cough with convulsions, in two infants aged 7 and 11 months; intestinal toxemia, in one aged 7 months; bronchopneumonia, in three aged 7, 14 and 18 months; acute peritonitis, in one aged 20 months; osteomyelitis, in one aged 2 years, and acute meningitis, in two aged 8 and 12 months. Karyorrhexis of the ganglion cells is also observed on the border of a small ischemic area in the nucleus caudatus of the brain of the last-mentioned infant. A marked glial reaction has occurred in the brain of the child aged 2 years with acute meningitis, and karyorrhexis of the ganglion cells is more pronounced than in the other brains of this group.

Karyorrhexis of astrocytes also occurs in the infant brain during the first two years of life. The affected cells are scattered in both the gray and the white substance, with more in the latter. Not many of them

are present in any of the brains, but they are more numerous in those of infants in the first six months of life. The older brains show a fairly constant number except in the presence of acute meningitis, in which karyorrhexis of astrocytes is somewhat increased in the cortex.

COMMENT

Since karyorrhexis of astrocytes is present, it may be thought that the changes which have been observed in the ganglion cells are those in an astrocyte superimposed on a ganglion cell. However, when the sections are well stained, the morphologic differences between karyorrhexis in the astrocyte and that in the ganglion cell are fairly distinct. In the ganglion cell the nuclear particles are larger, and the cytoplasm, although shrunken as in the astrocyte, still retains a recognizable pyramidal form. When the cytoplasm thins or is autolyzed in both cells by reason of the long time post mortem, the distinction depends on the size of the nuclear divisions. If these are slightly smaller than those usually found in the ganglion cell, the identity of the original cell is difficult to determine. This uncertainty is not encountered in the fading karyorrhexic astrocyte, as the nuclear divisions there are much finer than any ever seen in the ganglion cell, and the rounded outline of the cytoplasm occupies much less space.

Spatz [3] described karyorrhexis of the astrocyte in the spinal cord of the 5 day old rabbit. He believed that this change was physiologic, the cells probably having been worn out in aiding the production of myelin. This is possibly true of the karyorrhexic astrocyte in the infant brain, although karyorrhexic astrocytes are more numerous in the cortex before the beginning of myelination in the fourth month than after its completion in the seventh. Likewise they occured in the basal ganglions, where myelination is generally complete at birth. Spielmeyer [1] also thought that karyorrhexis of the glia cell is not always pathologic but may be an indication of the wear and tear of living; in lesions, the karyorrhexic astrocyte accompanies an increased growth of the glia. Dying astrocytes apparently attend the formation of new cells whether the stimulus to the development of the latter is physiologic or pathologic. Since the astrocytes of the infant brain are for the most part immature at birth and during the early postnatal months, when karyorrhexis is most prevalent, their destruction seems to be a reaction to the growth of young cells.

The significance of karyorrhexis of ganglion cells in the infant is perhaps comparable to that of karyorrhexis of astrocytes. The ganglion cells are also immature at birth and are not fully developed in the first

3. Spatz, H., in Nissl, F., and Alzheimer, A.: Histologie und Histopathologie. Arbeiten über die Grosshirnrinde, Jena, Gustav Fischer, 1918, vol. 6, p. 477.

half year, in which karyorrhexis chiefly occurs. Moreover, it is evident that this change differs from the descriptions given of cell injury in either infectious or "toxic" encephalitis. The nucleolus and the nuclear outline may be retained in the infectious encephalitic cell, but they are lost in the karyorrhexic ganglion cell of the infant brain. The nuclear particles of the latter cell are not fine or granular or confined within a nuclear periphery, as in the former, but are coarse and scattered. The cytoplasm is decreased rather than swollen, while absence of the tygroid character is the usual finding for young cortical cells. A faded cytoplasm and nuclear fragments are not a specific terminus, since such cells may indicate postmortem dissolution, while a hyperchromatic karyorrhexic ganglion cell in the infant may undergo neuronophagia by the microglia cell. Furthermore, destruction of the ganglion cell takes place in conditions which do not suggest cerebral lesions, as duodenal hematoma and tumor of the pharnyx. Immaturity of the cell is the dominant and common characteristic in all the cases. In the older brains, it is the more undeveloped second layer which harbors most of the few karyorrhexic ganglion cells. In similar diseases, as intestinal toxemia, a larger number of the disintegrating cells are found in the younger brains. Therefore, the greater maturity of the ganglion cells of the basal ganglions at birth perhaps accounts for the few karyorrhexic cells. As the Purkinje cells are also fairly large at birth and crowded, the nuclear particles found between them in the first month are probably those of disintegrating astrocytes.

Since karyorrhexis of the young ganglion and glia cells occurs regardless of the kind of fatal illness and probably in the absence of disease, it is possible that some of the destroyed cells are poorly formed or superfluous ones. However, the increase of karyorrhexis in the astrocytes of the cortex in acute meningitis is the expected reaction to this disease, as the glia cells are generally disturbed. But the greater number of karyorrhexic ganglion cells in the brain of the 2 year old child with acute meningitis is an exception, although the marked glial reaction indicates a severe infection. Intensity of the disease may explain, therefore, the observation that a few brains of the younger group show more pronounced destruction of the ganglion cells, even though the postmortem dissolution prevents recognition of all the affected cells. On the other hand, some young ganglion cells are perhaps less able than others to endure any pathologic disturbance during maturation, a sensitivity not equally shared by the astrocytes. Unusually poor resistance or immaturity may also be considered in relation to the karyorrhexic ganglion cells near the areas of vascular disturbance in the basal ganglions, since this is not a characteristic finding for ischemic parts in either the cortex or the basal ganglions.

It might be expected that gliosis would follow the destruction of the ganglion cells and that this would be apparent in brains of infants older than 6 months. However, so scattered are the karyorrhexic ganglion cells that not more than two astrocytes could replace the loss of one in a surrounding of ganglion cells, a gliosis which is not demonstrable. Furthermore, as the ganglion cells are close together at birth, the space of a single cell may be erased by the spread of the cells during the first two years. Although the lower part of the sixth layer has few cells in the first six months, a missing one easily escapes detection because during the first year this part of the layer is developed by the slow influx of ganglion cells from the subcortical white substance.

SUMMARY

A newly described change in the ganglion cells has been observed in the brains of thirty-four infants, 7 days to 2 years old. It is characterized by a break-up of the nuclear protoplasm into spherules and their dispersion in a shrunken, although sharply stained, cytoplasm, with terminal neuronophagia or disappearance by fading. The karyorrhexis is similar to that of the astrocytes which also occurs in the infant brain.

Since the karyorrhexic changes are found chiefly in the first six months of postnatal life, the immaturity of the ganglion and glia cells appears to be the factor predisposing toward their destruction by the antagonistic forces of growth. However, a variation in the number of disintegrating ganglion cells in brains from infants of similar age and disease seems to indicate either greater sensitivity in some of the cells or greater severity of the disease.

As only single, separated ganglion cells become karyorrhexic, any resulting gliosis is difficult to recognize in older cases.

TOOTH RING ANALYSIS

IV. NEONATAL DENTAL HYPOPLASIA: ANALYSIS OF THE TEETH OF AN INFANT WITH INJURY OF THE BRAIN AT BIRTH

ISAAC SCHOUR, D.D.S., PH.D.
AND
RUDOLF KRONFELD, M.D., D.D.S.
CHICAGO

INTRODUCTION [1] AND REVIEW OF THE LITERATURE

Birth is the most profound change in environment and nutrition which man experiences from conception to death. Therefore, it can reasonably be expected that birth and the immediately following neonatal period of adjustment will produce changes in many organs and tissues. But no matter how marked these changes may have been, they soon become indistinct and finally completely obliterated by the continuous process of growth and replacement to which most tissues are subjected.

The teeth, however, are a noteworthy exception to this. Changes effected during the growth and calcification of the dentin and enamel remain permanently engraved on these structures. They can be recognized at any later time, regardless of whether the tooth is still in the mouth or whether it has been shed or extracted. Neonatal injuries are observed in deciduous teeth and occasionally in that portion of the first permanent molar which was growing and calcifying at the time of birth.

The idea that birth with its associated nutritional and metabolic disturbances may leave its permanent marks on the teeth is rather new. Only during the past decade have hypoplastic defects in the deciduous teeth received much attention from dentists and pediatricians. The lack of knowledge concerning the growth rate of dentin and enamel and also the apparent difficulty of finding a landmark in these structures which would identify the time of birth made it impossible to ascribe such defects definitely to a certain period of life.

From the Department of Histology, College of Dentistry, University of Illinois, and the Foundation for Dental Research of the Chicago College of Dental Surgery.

1. The preceding studies on tooth ring analysis have been reported by: (*a*) Schour, I., and Steadman, S. R.: Anat. Rec. **63**:325, 1935. (*b*) Schour, I.: J. Am. Dent. A. **23**:1946, 1936. (*c*) Schour, I., and Poncher, H. G.: Am. J. Dis. Child. **54**:757, 1937.

One of the earliest attempts to distinguish histologically between prenatal and postnatal dental hard tissues was made by Karnosh [2] in his analysis of syphilitic hypoplasia of the enamel. In ground sections of hypoplastic first permanent molars of patients with congenital syphilis ("mulberry molars"), he found a thin zone over the dento-enamel junction, at the tips of one or several cusps, in which the enamel appeared normal. He termed this zone the "prenatal cap of enamel." Above the prenatal enamel an irregular zone of hypoplastic enamel was present, which Karnosh described as the "hypoplastic first-year layer." He recognized clearly the connection between hypoplasia and disturbances occurring at birth, as indicated by a statement in the conclusion of his paper: "A time factor of congenital syphilis as a cause of dental hypoplasia seems to be established in the first year of life, the effects coming on suddenly at birth and being concurrent with the stomatitis, rhinitis and the other familiar symptoms of syphilis in the new-born."

Rushton [3] reported the usual occurrence of a fine contour line in the enamel formed at the time of birth and described it as the "birth line."

Stein,[4] in his paper on enamel defects in deciduous teeth, pointed out that these defects are frequently located along the line where the prenatal and the postnatal portions of the tooth meet. He emphasized the "shock" nature of the birth process and the possibility that such a shock may well result in disturbed calcium metabolism and in poorly formed enamel. He showed excellent illustrations of ground sections in which a dividing line between the older intact and the newer hypoplastic enamel could be clearly seen, but he was not in a position to establish the exact significance of this dividing line.

In 1936 one of us (I. S.),[1b] without knowledge of Rushton's work,[3] reported the finding of the neonatal ring in the human deciduous teeth and in the first permanent molar. This ring is a pronounced stria of Retzius in the enamel and a corresponding contour line in the dentin; it is the result of a disturbance of growth and calcification at birth and shortly thereafter. This line in the enamel and dentin was established as a neonatal ring on the following basis: (1) the findings in the teeth of an infant which at known intervals had received injections of a substance which produced marks in the enamel and dentin,[1c] (2) the recognition that human enamel and dentin grow at an average daily rate of 4 microns, with a growth gradient that is characteristic and

2. Karnosh, L. J.: Arch. Dermat. & Syph. **13**:25, 1926.

3. Rushton, M. A.: Dent. Rec. **53**:170, 1933.

4. Stein, G.: Ztschr. f. Stomatol. **34**:843, 1936.

constant for any particular level of a given tooth class,[5] (3) the identity of the surface of the enamel and dentin in the deciduous teeth in full term stillborn infants as compared with the position and level of the neonatal ring in the teeth of older infants.[1b] Equipped with this knowledge, one may now find the neonatal ring in almost every human deciduous tooth and may determine the amount of enamel and dentin that was laid down before and after birth. The amount laid down before birth can be found by measuring the distance from the neonatal rings in the enamel and dentin to the dento-enamel junction. The amount laid down after birth is indicated by the distance from the neonatal rings to the enamel or pulpal surfaces. The measurements are made along the direction of the enamel rods or dentinal tubules, which represent the path of growth.

In human deciduous teeth enamel is being laid down at an average rate of 4 microns in twenty-four hours.[5] If in a given tooth the greatest thickness of the enamel between the dento-enamel junction and the neonatal ring is 400 microns, and the thickness between the neonatal ring and the enamel surface is 640 microns, it can be concluded that in this particular tooth the enamel began to form about one hundred days before birth and that enamel formation continued for about one hundred and sixty days after birth.

It is obvious that the greater the disturbance at birth, the more accentuated will be the neonatal ring. *The more difficult it is for the organism to make the adjustment from the sheltered intrauterine life to the independent postnatal existence, the more pronounced will be the zone of poorly formed and calcified enamel and dentin apposed during this period.* It may also be expected, as indicated by Stein,[4] that the neonatal ring will be more prominent in the teeth of prematurely born children than in infants born at term, because of the relatively greater nutritional difficulties and other disturbances usually encountered by premature infants.

The present report is based on the findings in the jaws and teeth of a colored infant who suffered an injury of the brain at birth from which it never recovered and which was a contributing cause of its death at the age of 7 months and 5 days. The severity of the trauma, coupled with the clinical history, makes the study of the dental conditions particularly interesting.

HISTORY OF CASE

A Negro girl, born at term, with a normal delivery, weighed at birth 2.8 Kg. She appeared to be well until the age of 3 months, when she became irritable and at times apathetic. She showed attacks of muscular rigidity, which began in the arms, spread to the legs and finally involved the whole body. She was unable to hold up her head.

5. Schour, I., and Massler, M.: J. Dent. Research **16**:349, 1937.

This patient was brought to the hospital with an infection of the upper respiratory tract. She was undernourished, poorly developed and listless. The fontanels were patent. The pupils reacted to light but did not focus on objects. All extremities were spastic, with hyperactive deep tendon reflexes.

The clinical diagnosis on admission was: injury of the brain, sustained at birth; spastic panplegia; blindness; infection of the upper respiratory tract. Tests for syphilis were negative; the white blood cells numbered 14,200.

Bronchopneumonia developed, to which the child succumbed at the age of 7 months and 5 days (218 days).

Autopsy.—The body was that of a fairly well developed, poorly nourished Negro girl. It weighed 4 Kg. and was 58 cm. long. The conditions noted were general malnutrition, focal bronchopneumonia in the right lung and parenchymatous degeneration of the heart muscle, liver, kidneys and spleen.

The changes in the brain were indicative of a trauma occurring at birth. In many areas the number of ganglion cells was reduced, and there was an increase in the glia cells. These changes were particularly prominent in the region of the basal ganglions. Throughout the brain the capillaries were engorged and prominent, and the walls of the larger vessels were thickened.

HISTOLOGIC PREPARATIONS

The jaws were removed at autopsy and fixed in 4 per cent formaldehyde and 95 per cent alcohol. Roentgenograms were made of both jaws. The jaws were then divided in the midline, and one half of each upper and lower jaw was decalcified, embedded in pyroxylin, sectioned and stained with hematoxylin and eosin, a modified Rio Hortega silver stain and Mallory's triple connective tissue stain. The other half was first embedded in thick pyroxylin, without previous decalcification, and then the pyroxylin block was embedded in plaster. Labiolingual or buccolingual slabs of the jaw, each about 2 mm. thick, were cut with a circular saw of superspeed steel, after which the slabs were ground on Black's grinding machine to a thickness of about 20 microns. In this way it was possible to compare decalcified stained sections and ground sections of corresponding teeth of this infant. This comparison was particularly important in the study of the enamel.

ROENTGENOGRAMS

The roentgenograms showed a state of calcification which corresponds to that found in a child approximately 6 months old. In the deciduous cuspids and molars there were evidences of hypoplasia of the enamel in the portion of the tooth formed and calcified at birth (fig. 1). Calcification had begun in the permanent central incisors, lateral incisors and cuspids, except for the upper lateral incisors, and in the first permanent molars, in which four or five individual calcified growth centers were visible.

ENAMEL OF DECIDUOUS TEETH

Neonatal Ring in the Enamel.—The study of the neonatal rings in the enamel of the deciduous teeth was made largely on ground sections, since most of the enamel had been dissolved from the decalcified

sections. The neonatal ring appeared as a dark line in the enamel, dividing the prenatal and the postnatal portion. The neonatal injury to the enamel was much more severe than that to the dentin. Whereas the disturbance of the dentin formation was only temporary and was apparently followed by complete recovery in most of the dentin-forming cells, the enamel formation, with few exceptions, was permanently arrested at birth, so that in almost every deciduous tooth the enamel surface and the neonatal ring coincided.

Postnatal Enamel Formation.—Amelogenesis continued past the neonatal ring only in the lower deciduous cuspids and the second deciduous molars. In these teeth an irregular amount of postnatal enamel had been formed over the neonatal ring (fig. 2), which appeared as a dark line in the enamel, dividing the prenatal and postnatal layers. Under high magnification, the enamel rods could be seen not only changing in color but deviating in their course (fig. 4, *3*).

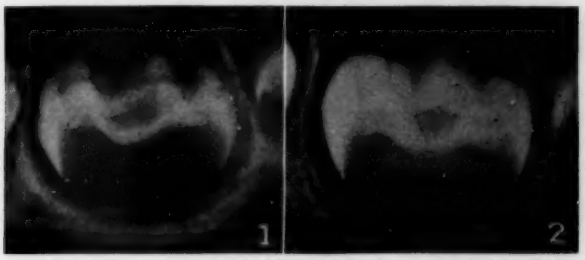


Fig. 1.—Roentgenogram of the lower second deciduous molar of the jaw reported on here (*1*) and of the lower second deciduous molar of another jaw of approximately the same chronologic age but without hypoplasia of the enamel (*2*). In length and height the two tooth germs are nearly equal; the thickness of the calcified portion in *1*, however, is greatly reduced by hypoplastic defects of the enamel, and the cuspal outline appears irregular and abnormal.

Gingivally to the neonatal portion of the tooth, postnatal enamel had been laid down in the form of a circular ledge. This enamel was formed by ameloblasts which had differentiated and started to function after the neonatal arrest of growth (fig. 3, *3* and fig. 5, *3*). This circular postnatal enamel ledge, in contrast to the absence of enamel immediately occlusal to its level, appeared to be very prominent and suggested the possibility of compensatory hyperplasia of the enamel. Micrometer measurements, however, showed no increased thickness in the enamel. Figure 2 shows that the curvature of the surface of the gingival enamel ledge, except for the portion immediately adjacent to the hypoplastic pit, corresponded with the general curvature of the enamel surface that would have existed if the hypoplasia had not occurred.

Enamel Epithelium.—The condition of the enamel epithelium varied in different levels and teeth. In some areas there was no epithelium, and the enamel was covered by connective tissue. In other areas, usually above the cusps, stratified squamous epithelium from three to twelve layers in thickness covered the enamel. Degenerative changes were most common in the gingival portion and consisted of a ballooning of the ameloblasts, similar to the condition described by Bauer [6] and by Klein.[7]

DENTIN OF DECIDUOUS TEETH

Prenatal Dentin.—The neonatal ring was clearly visible in the dentin of every deciduous tooth. A comparison of the conditions of calcifi-

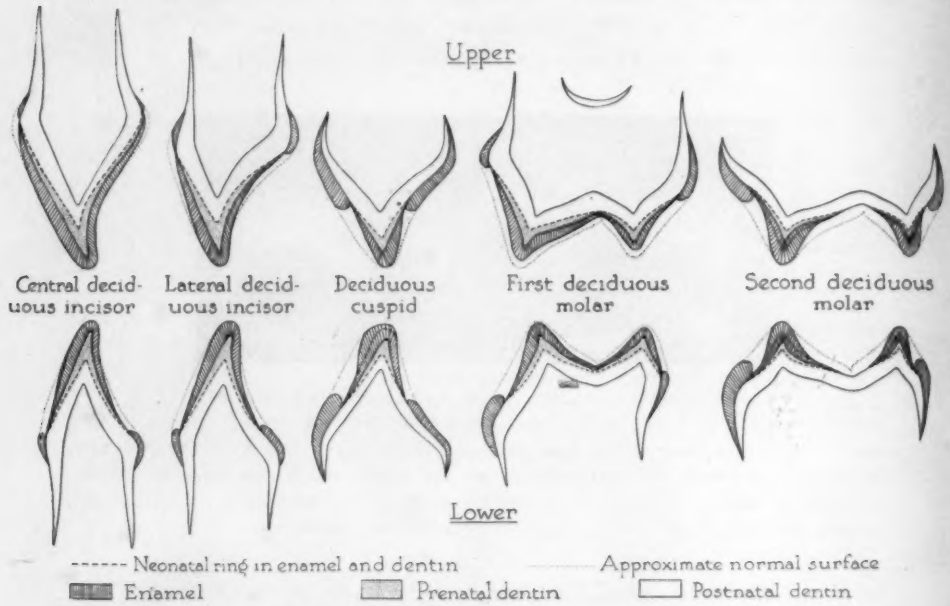


Fig. 2.—Semidiagrammatic tracings of the hypoplastic upper and lower deciduous teeth. Note that the formation of dentin has continued in all teeth after birth, whereas the formation of enamel has in most instances terminated at birth. Postnatal enamel was laid down in the cervical portion of the crown, where the ameloblasts were not yet active at the time of birth; only in the lower deciduous cuspid and in the second deciduous molars has postnatal enamel been formed on top of the prenatal enamel layer.

cation in prenatal and postnatal dentin is interesting. We tried to evaluate the degree of calcification on the basis of the staining proper-

6. Bauer, W.: Vrtljsschr. f. Zahnh. **45**:62, 1929.
7. Klein, H.: J. Am. Dent. A. **18**:866, 1932.

ties, in accordance with the thesis that the more highly calcified a structure is (before decalcification in acid) the greater affinity it will have for hematoxylin. Thus, we found that the prenatal dentin was more uniformly and more intensively calcified than the postnatal dentin (fig. 3, *2*).

Neonatal Ring in the Dentin.—Between the prenatal and the postnatal dentin lay a narrow zone of light (eosin-stained) dentin, which represented the predentin present at the time of birth (fig. 3, *2*, Pd_1). This predentin, however, never became calcified and apparently remained in the condition which it had attained at the time of the birth trauma. Its staining reaction was the same as that of the predentin next to the pulp, both in sections stained with Mallory's triple connective tissue stain and in sections stained with hematoxylin and eosin.

The location of the neonatal ring with reference to the dento-enamel junction varied in the different teeth; it was slightly above the level of the normal full term position described in a previous report (Schour [1b]). This may indicate that the infant was slightly premature, which assumption is further corroborated by the slightly subnormal weight at birth.

Neonatal Dentinal Plexus.—The study of decalcified sections stained with silver and of unstained ground sections revealed a heretofore unknown behavior of the dentinal tubules at the neonatal ring (fig. 5, *2* and *4*). The tubules in the prenatal dentin were normal in course and number. But there was a definite and sudden change in the course, number and diameter of the tubules at the junction between the prenatal and the postnatal dentin. The periphery of the postnatal dentin gave the appearance of extensive terminal branching of the tubules, similar to the normal terminal plexus at the dento-enamel junction.

This neonatal dentinal plexus was the result of a fusion of several prenatal tubules into one wider postnatal tubule at the neonatal ring. The number of tubules in the postnatal dentin was approximately one third of the number in the prenatal dentin (fig. 5, *2*). A small proportion of the tubules of the prenatal dentin continued in the postnatal dentin, but a greater number bent and joined adjacent tubules at the beginning of the postnatal dentin. In the latter the dentinal tubules were thicker in diameter.

A sharp deflection from the normal direction of the dentinal tubules was seen at the gingival portion of the neonatal ring (fig. 3, *4*). This disturbance in the course and branching of the dentinal tubules closely resembled the changes, reported by Orban,[8] that result from sudden mechanical or chemical insults to the formative cells of the dentin. The resultant line may be termed "a line of deviated growth."

8. Orban, B.: Ztschr. f. Stomatol. **32**:303, 388, 434 and 504, 1934.

EXPLANATION OF FIGURE 3

1, photomicrograph of a labiolingual section through the lower right deciduous and permanent lateral incisors; × 5. *J* indicates the jaw ridge; *E* designates the enamel, *D* the dentin and *P* the pulp of a deciduous incisor. *Pg* indicates the tooth germ of a permanent incisor; *M*, marrow spaces in the body of the mandible.

2, higher magnification of area *2* in *1*; × 84. *F* indicates connective tissue of a tooth follicle; Pd_1, predentin present at the time of birth; *NR*, the neonatal ring in the dentin; *Ig*, interglobular dentin; Pd_2, predentin present at the time of death; *Od*, odontoblasts; *P*, pulp.

3, photomicrograph of a labiolingual ground section through the lower left deciduous lateral incisor; × 10. E_1 indicates prenatal enamel; E_2, postnatal enamel; *D*, dentin; *N*, the neonatal ring in the dentin.

4, higher magnification of area 4 in *3*; × 82. *DEJ* indicates dento-enamel junction; Pd_1, predentin present at the time of birth; *NR*, the neonatal ring in the dentin; *NP*, the neonatal plexus of the dentinal tubules; *X*, an area in which postnatal enamel failed to form.

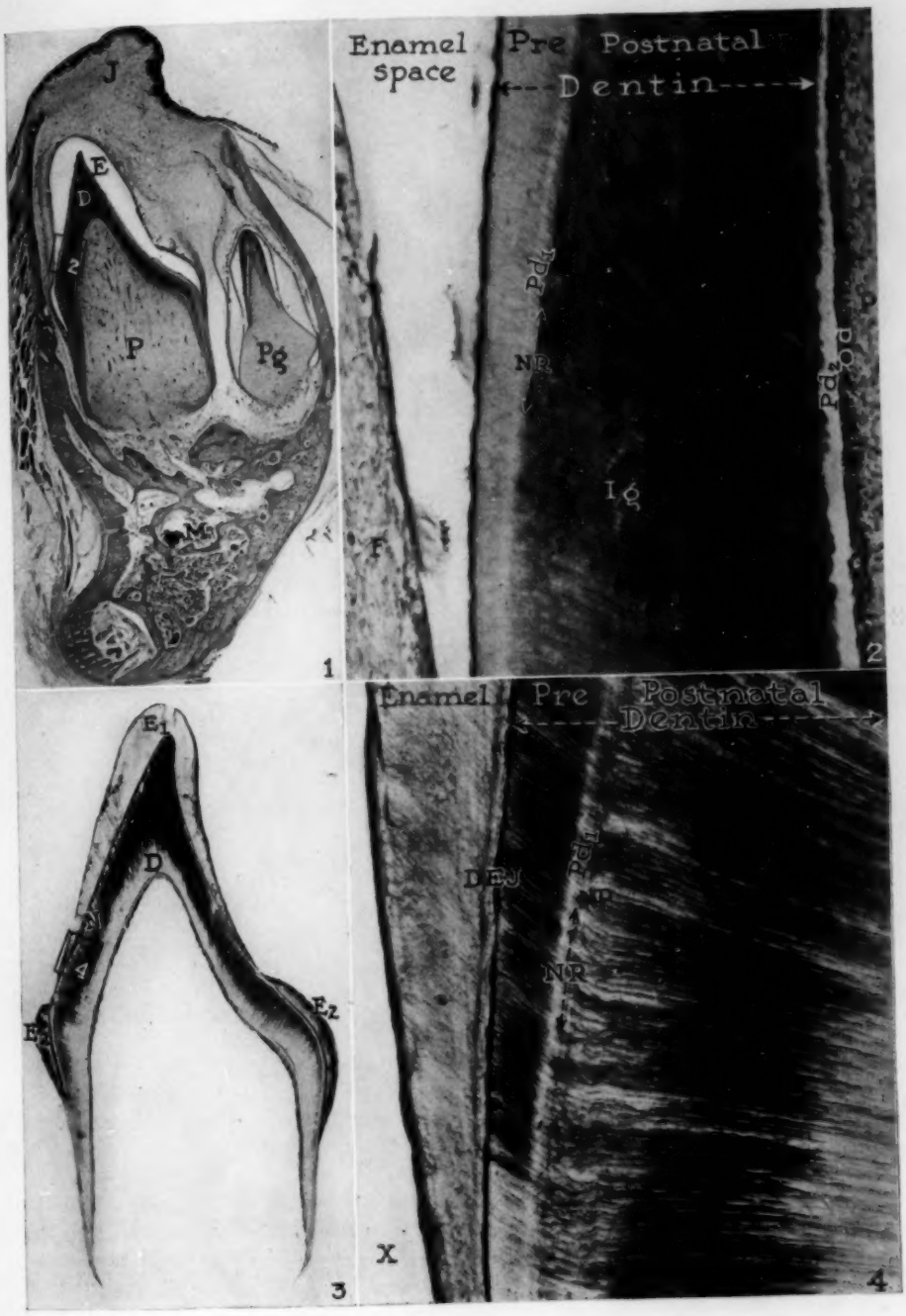

J
E
D
2
P
Pg
M
F
1
Enamel space
Pre
Postnatal
Dentin
Pd1
NR
Ig
Pd2od
2
E1
D
4
E2
3
Enamel
Pre
Postnatal
Dentin
DEJ
Pd1
NR
X
4

Figure 3

The neonatal zone in the dentin of these teeth, therefore, showed the following features: (1) an uncalcified zone, appearing light in the sections stained with hematoxylin and eosin, which was the predentin present at birth; (2) a hypercalcific zone appearing dark in the sections

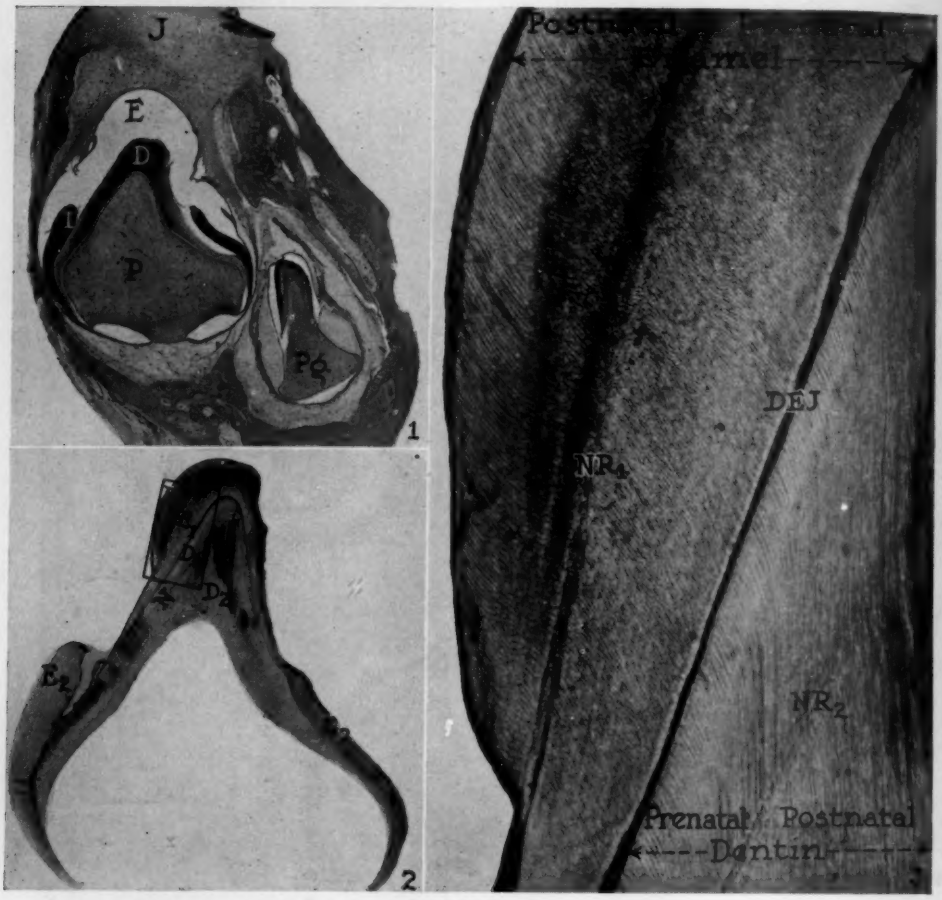


Figure 4

1, photomicrograph of a labiolingual section through the lower right deciduous and permanent cuspids; × 5. *J* indicates the jaw ridge; *E* designates the enamel space, *I* the immature enamel, *D* the dentin and *P* the pulp of a deciduous cuspid. *Pg* indicates the tooth germ of a permanent cuspid.

2, photomicrograph of a labiolingual ground section through the lower left deciduous cuspid; × 10. E_1 indicates prenatal enamel; E_2, postnatal enamel; D_1, prenatal dentin; *N*, the neonatal ring in the dentin; D_2, postnatal dentin.

3, higher magnification of the area indicated in *2*; × 91. NR_1 indicates the neonatal ring in the enamel; *DEJ*, the dento-enamel junction; NR_2, the neonatal ring in the dentin.

stained with hematoxylin and eosin, corresponding to the outermost layer of the postnatal dentin; (3) a definite deviation and change in the number of the dentinal tubules as they passed from the prenatal dentin into the postnatal dentin, and (4) the formation of a "neonatal dentinal plexus" at the periphery of the postnatal dentin.

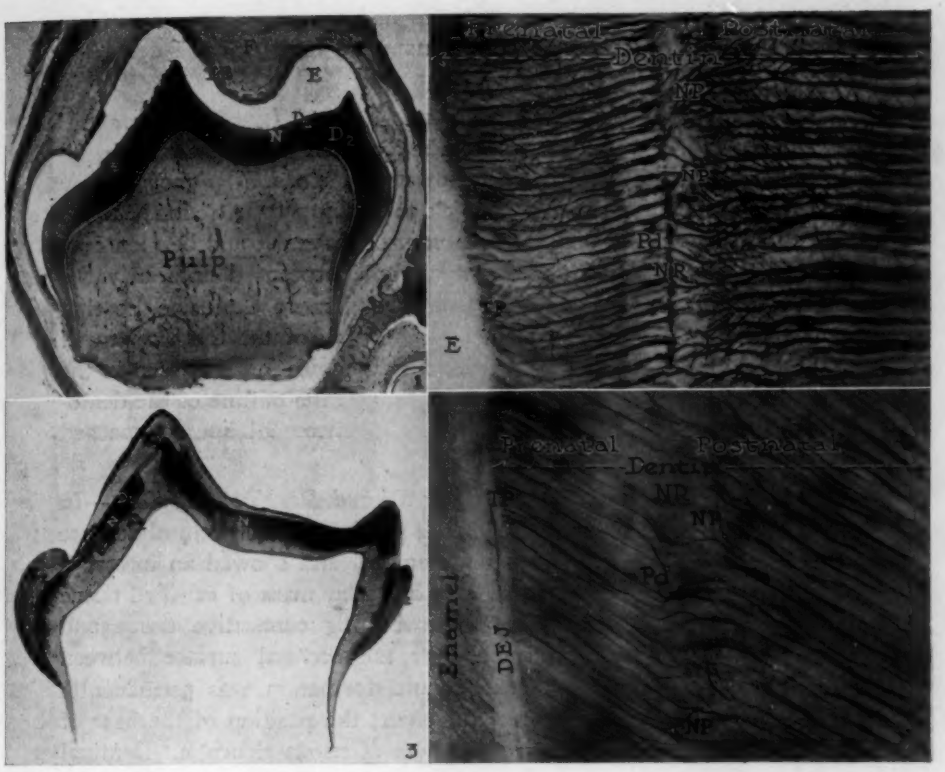


Figure 5

1, photomicrograph of a buccolingual section through the right lower first deciduous molar; × 11. *F* indicates connective tissue of a tooth follicle; *EE*, enamel epithelium; *E*, enamel space; D_1, prenatal dentin; *N*, the neonatal ring in the dentin; D_2, postnatal dentin.

2, higher magnification of area 2 in *1;* tannin-silver stain after Rio Hortega. *E* indicates enamel space; *TP*, a terminal plexus of dentinal tubules at the dento-enamel junction; *Pd*, predentin present at birth; *NR*, the neonatal ring in the dentin; *NP*, a neonatal plexus of the dentinal tubules.

3, photomicrograph of buccolingual ground section through the lower left first deciduous molar; × 11. E_1 indicates prenatal enamel; E_2, postnatal enamel; D_1, prenatal dentin; *N*, the neonatal ring in the dentin; D_2, postnatal dentin.

4, higher magnification of area 4 in *3*. *DEJ* indicates the dento-enamel junction; *TP*, a terminal plexus of dentinal tubules at the dento-enamel junction; *Pd*, predentin present at birth; *NR*, the neonatal ring in the dentin; *NP*, a neonatal plexus of the dentinal tubules.

Postnatal Dentin.—The dentin formed after birth showed irregular, cloudy calcification, with numerous areas of uncalcified interglobular dentin (fig. 3, *2, Ig*). It was separated from the prenatal predentin by a very dark hypercalcified line. In the figure, Pd_3 indicates the predentin which was laid down just before death and which was not yet calcified. An irregular border separated the postnatal dentin and the predentin adjacent to the pulp. The odontoblasts and the pulp showed nothing unusual. Identical pathologic conditions were found in the dentin of every deciduous tooth (fig. 6, *2*).

Changes in the Outline of the Dento-Enamel Junction Associated with the Neonatal Injury.—Where the neonatal ring met the dento-enamel junction, the latter often showed a distortion in morphologic outline that ranged from a slight nick to a sharp constriction. In the posterior teeth such distortions were very slight and could be seen only under high magnification. However, as one proceeded anteriorly the constrictions became increasingly greater and more extensive, so that in the upper deciduous central incisor the neck of the tooth appeared to be markedly constricted. A corresponding deviation was seen in the pulpal outline (fig. 2). The neonatal effect on the outline of the dento-enamel junction appeared to increase as the neonatal line approached closer to the cemento-enamel junction.

Neonatal Odontoceles in the Upper Second Deciduous Molars.—In each upper second deciduous molar, the portion of the occlusal surface within the sulcus was not covered by enamel and showed an interruption of the dento-enamel junction. An irregular mass of calcified tissue protruded from the dentin into the overlying connective tissue and spread laterally like a mushroom over the occlusal surface between the cusps (fig. 6, *1* and *3*). The enamel formation was permanently arrested in this region at the time of birth; the position of the base of the odontocele showed that the latter was of neonatal origin. Dentinal tubules projected into this mass, which consisted for the most part of a bonelike structure arranged about nutrient vessels.

Neonatal Arrest of Growth.—The amount of postnatal dentin was less than normally would be expected in a 7 month old infant. The dentin development seemed to be about two months late. This was demonstrated by micrometer measurements. Table 1 shows the amount of postnatal dentin formed in each tooth and the estimated duration of the neonatal arrest in the formation of dentin. The neonatal arrest of growth, which normally lasts about two weeks, in this case was prolonged to two months because of the injury at birth. The average period of neonatal arrest of all the deciduous teeth was 71.2 days (average of all figures in the last column of table 1).

NEONATAL HYPOPLASIA OF THE LOWER LEFT FIRST PERMANENT MOLAR

In the lower first permanent molars the formation of enamel and dentin had begun in the mesiobuccal cusps just before birth. The neonatal ring was evident only in that location. The thickness of the prenatally formed dentin was 50 microns and that of the prenatally formed enamel 48 microns; this indicates that hard tissue had begun to form about twelve days before birth (fig. *7, 3*).

In the lower left first permanent molar, the formation of enamel had become permanently arrested over the prenatal cone, so that a small pit-shaped area of hypoplasia of the enamel resulted at the tip of the mesiobuccal cusp (fig. *7, 1* and *2*). The prenatal enamel cone took a deep hematoxylin stain, a greenish yellow color with the silver stain and a blue color with Mallory's triple connective tissue stain.

TABLE 1.—*The Amount of Postnatal Dentin Formed and the Calculated Duration of Its Formation in Days*

Teeth Examined*	Distance from Tip of Neonatal Dentinal Ring to Tip of Pulpal Horn, Microns	Average Rate of Apposition for This Region per 24 Hours, Microns†	Calculated Number of Days of Active Postnatal Formation of Dentin	Calculated Number of Days of Neonatal Arrest in Apposition‡
Deciduous central incisors.....	1,080	8	135	82
Deciduous lateral incisors.....	1,075	7.5	143.3	74.7
Deciduous cuspids.....	950	6	158.3	59.7
Deciduous first molars.....	840	5.5	152.7	66.0
Deciduous second molars.....	800	5.5	145.4	72.6

* The averages of the upper and lower teeth are considered together. A slight difference exists between the upper and the lower teeth.

† The average rate of growth for different regions of the deciduous teeth as given by Schour and Massler.[5]

‡ The average period of neonatal arrest for all the deciduous teeth was 71.2 days.

The dentinal tubules in the lower first permanent molars showed the same changes in their course and number at the neonatal ring as have been described for the deciduous teeth. The neonatal predentin had failed to become calcified and could be clearly recognized by its light color (fig. *7, 3, Pd*). Pulpward from the neonatal line a narrow hypercalcific layer represented the first dentin laid down after birth.

Odontomes Near the First Permanent Molar.—In the connective tissue which overlay the occlusal surface of the lower right first permanent molar were two small circumscribed odontomes, which consisted of concentrically arranged layers of enamel and dentin in a state of active apposition, surrounding a formative papilla (fig. *7, 4*).

CHRONOLOGY OF DECIDUOUS TEETH

To determine the time of the beginning of dentin formation in the deciduous teeth, measurements were made in the dentin from the highest point on the neonatal ring to the corresponding growth center on the

EXPLANATION OF FIGURE 6

1, photomicrograph of a buccolingual section through the upper right second deciduous molar; × 11. *F* indicates connective tissue of a tooth follicle; E_1, enamel space; E_2, postnatal enamel; *D*, dentin; *O*, an odontocele; *HS*, Hertwig's sheath.

2, higher magnification of area 2 in *1*; × 83. *A* indicates ameloblasts; E_2, postnatal enamel; E_1, a thin layer of prenatal enamel; D_1, prenatal dentin; *NR*, the neonatal ring in the dentin; D_2, postnatal dentin; *Ig*, interglobular dentin; *Pd*, predentin.

3, higher magnification of area 3 in *1*; × 56. D_1 indicates prenatal dentin; *NR*, the neonatal ring in the dentin; D_2, postnatal dentin; *Pd*, predentin; D_3, irregular dentin in the odontocele; *C*, fibrous calcified material resembling cementum or bone.

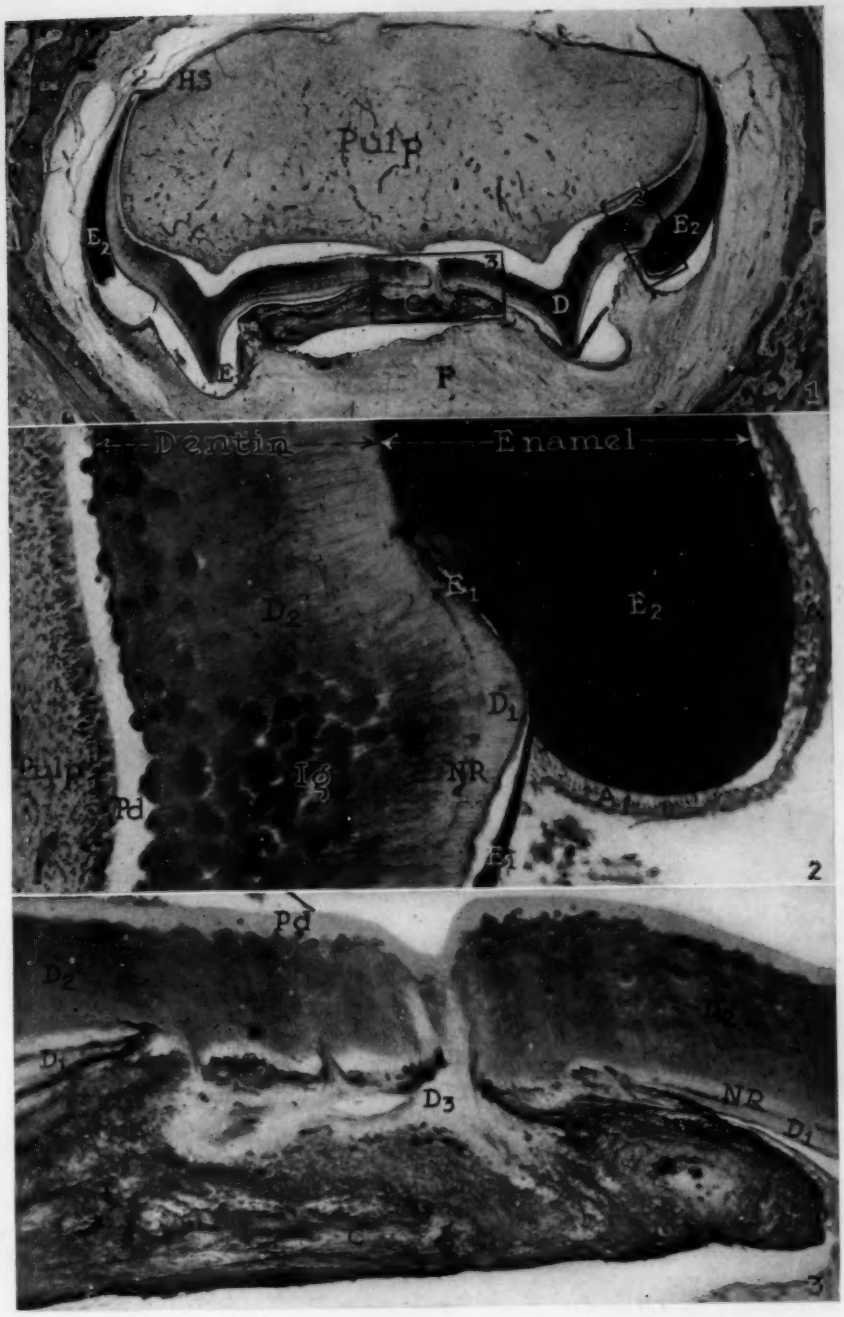

HS
Pulp
E2
E2
D
E
F
1
Dentin
Enamel
E1
E2
D2
D1
Ig
NR
Pd
E1
2
Pd
D2
D1
D3
D2
NR
D1
3

Figure 6

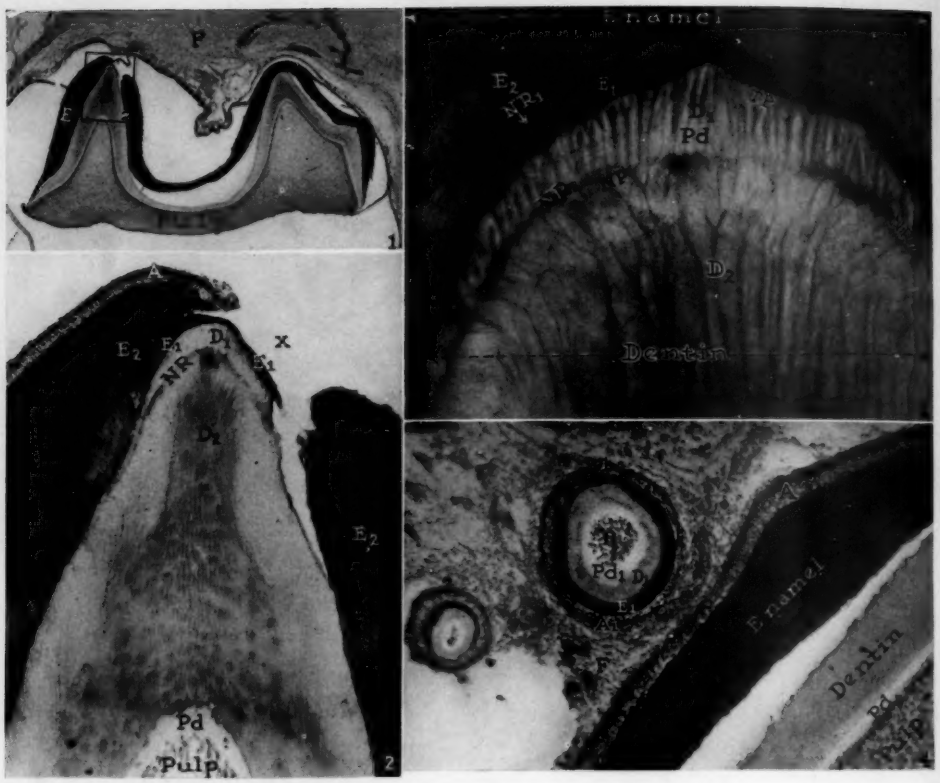


Figure 7

1, photomicrograph of a buccolingual section through the right lower first permanent molar; × 7. *F* indicates connective tissue of a tooth follicle; *E* designates the enamel, and *D* the dentin of the first permanent molar.

2, higher magnification of area 2 in *1*; × 63. *A* indicates ameloblasts; E_1, prenatal enamel; E_2, postnatal enamel; D_1, prenatal dentin; *NR*, the neonatal ring in the dentin; D_2, postnatal dentin; *Pd*, predentin; *X*, a hypoplastic defect in the postnatal enamel.

3, high magnification of the tip of the cusp corresponding to *2*; × 252.5. E_2 indicates postnatal enamel; NR_1, the neonatal ring in the enamel; E_1, prenatal enamel; D_1, prenatal dentin; *TP*, a terminal plexus of the dentinal tubules at the dento-enamel junction; *Pd*, predentin present at time of birth; NR_2, the neonatal ring in the dentin; D_2, postnatal dentin; *NP*, a neonatal plexus of the dentinal tubules.

4, two small odontomes (*1* and *2*) in the connective tissue overlying the crown of the lower first permanent molar; × 37.5. *A* indicates ameloblasts and *Pd* predentin of the first permanent molar, and *F*, a tooth follicle. *A* indicates the ameloblasts, E_1, the enamel, D_1 the dentin, Pd_1 the predentin and P_1, the pulp of the odontome.

dento-enamel junction. The average rate of postnatal formation of dentin in this region (table 1) was taken to approximate the average rate of prenatal formation. This is valid in view of the fact that the range of variability even under extreme circumstances would be less than twenty days. The onset and the course of prenatal formation, of course, were not affected by the neonatal injury. The results are given in table 2 and may be taken as representing average normal values.

CHRONOLOGY OF PERMANENT TEETH

With the possible exception of that in the first permanent molar, the formation of hard tissue in the permanent teeth does not begin during the neonatal period. Consequently the beginning of the formation of the

TABLE 2.—*Beginning of Dentin Formation in the Deciduous Teeth*

Deciduous Teeth	Upper	Lower
Central incisor	4 mo. in utero	4½ mo. in utero
Lateral incisor	4½ mo. in utero	4½ mo. in utero
Cuspid	5 mo. in utero	5 mo. in utero
First molar	5 mo. in utero	5 mo. in utero
Second molar	5½ mo. in utero	5½ mo. in utero

TABLE 3.—*Beginning of the Formation of the Dentin of the Permanent Teeth*

Permanent Teeth	Upper	Lower
Central incisor	2-3 mo.	3 mo.
Lateral incisor	Not yet begun	3 mo.
Cuspid	4 mo.	4 mo.
First molar	2 weeks	Mesiobuccal cusp, 12 days before birth; other cusps, 2 weeks after birth

hard tissue of these teeth had not been affected by the neonatal injury and represented the normal onset of calcification of the permanent teeth. In all the germs of the permanent teeth, the greatest distance from the tips of the pulp horns to the dento-enamel junction was measured. The average rate of dentin formation was estimated to range from 5 microns in the permanent central incisors to 4 microns in the permanent cuspids. These averages are approximate and await confirmation by experimental studies but are valid in this particular case, since the range of variability is less than twenty-five days. The onset of formation of the permanent teeth is given in table 3.

These figures are an additional confirmation of the work of Logan and Kronfeld [9] and of Kronfeld [10] on the development of the permanent

9. Logan, W. H. G., and Kronfeld, R.: J. Am. Dent. A. **20**:379, 1933.
10. Kronfeld, R.: J. Am. Dent. A. **22**:1131, 1935.

dentition. These authors showed that the calcification of the permanent anterior teeth takes place considerably earlier than was generally believed prior to their reports. This is especially true of the permanent cuspids, which were found to develop almost simultaneously with the central incisors.[11] A too early date, on the other hand, was usually given for the onset of calcification of the first permanent molars, until it was found that these teeth begin to calcify about the time of birth.[10] The measurements in this jaw with a known clinical history make it possible to determine the onset of calcification more accurately than was ever possible before, and we were rather surprised to find still earlier ages for the beginning of formation and calcification than those reported in the latest publications on this subject by one of us (R. K.).

COMMENT

Dental Hypoplasia.—Completed enamel is like a tombstone, for on it is inscribed the history of the vicissitudes of the ameloblasts. Completed dentin, like enamel, though to a lesser extent, constitutes a permanent record of the systemic and local conditions which influenced the cellular factors responsible for the formation of dentin. The hypoplastic defect is an end product, and *its genesis is ultimately cellular.* In this comment, therefore, we shall attempt to trace the possible cytogenesis of dental hypoplasia.

Since a direct relationship between an injury to the brain and the formation of a tooth cannot be readily assumed, it may be suggested that the neonatal injury to the brain resulted primarily in a prolonged state of malnutrition, which in turn affected the formative cells of the teeth. Infants suffering from such injuries of the brain are apathetic, unresponsive and weak. Often they are extremely undernourished.

(*a*) Enamel: The ameloblasts which at the time of the injury at birth were actively engaged in the formation of enamel suffered most severely. In the majority of the teeth amelogenesis was permanently arrested at birth. Occasionally, however, recovery of the ameloblasts occurred (fig. 4, *2* and *3*). On the other hand, the cells of the enamel organ which at the time of birth were still in their differentiating stage preparatory to the formation of enamel were unaffected by the injury. The result was a circular ledge of postnatal enamel adjacent to a hypoplastic area composed only of prenatal enamel (figs. 3, 4, 5 and 6). The level and contour of this hypoplasia correspond to the erstwhile position of those ameloblasts present at the time of birth and, therefore, to the position of the neonatal ring.

The selective reactivity of the ameloblasts depending on the particular stage of their life cycle is in harmony with what is known

11. Kronfeld, R.: J. Am. Dent. A. **22**:1521, 1935.

regarding the factors influencing the reaction of an organ to given interferences (Gudernatsch [12]).

(*b*) Dentin: The findings in the dentin indicate that the odontoblasts functioning at the time of birth responded to the neonatal shock, like the ameloblasts, by a complete cessation of activity. However, a much greater number of dentin-forming cells than of enamel-forming cells recovered after the seventy day arrest of growth. The sudden decrease in the number of postnatal dentinal tubules suggests that approximately one third of the odontoblasts recovered. Apparently the dentin-forming connective tissue cells are less vulnerable than the more highly specialized enamel-forming epithelial cells. This is in harmony with the reaction of ameloblasts and odontoblasts to sodium fluoride.[1c]

Odontocele.—The position of an odontocele, a rare hyperplastic malformation, in the occlusal surface of a second deciduous molar indicates definitely its neonatal origin (fig. 6, *1* and *3*). This type of malformation—a protrusion of a mushroom-like, irregularly calcified structure from the center of the crown of a molar—is described in the literature by Herbst and Apffelstaedt,[13] who termed it odontocele. They suggested that it may be of traumatic origin, resulting from an extrusion of dentinogenic cells through the uncalcified area between the calcified cusps at the time of birth, presumably through pressure on the head and jaws during a delivery by forceps.

Possible Explanation for the Formation of Odontomes.—The presence of the small odontomes (fig. 7, *4*) near the occlusal surface of the permanent first molars suggests the possibility that they, like the odontocele, are associated with the neonatal trauma. It is conceivable that as a result of the injury at birth a small portion of the enamel organ of the first permanent molar, which was just beginning to enter the appositional stage of its life cycle, was pinched off and became displaced in the overlying connective tissue. There it continued to develop and formed enamel, dentin and pulp.

SUMMARY

The teeth of a human infant 218 days of age that had suffered an injury of the brain at birth were studied in roentgenograms and in ground and decalcified histologic sections.

The neonatal ring in enamel and dentin, which is normally a manifestation of the physiologic neonatal arrest of growth, was found to be pathologically accentuated. Micrometer measurements indicated a neo-

12. Gudernatsch, F.: Specific Chemical Factors Influencing Growth and Differentiation, in Cold Spring Harbor Symposia on Quantitative Biology, Cold Spring Harbor, L. I., New York, The Biological Laboratory, 1934, vol. 2, p. 94.

13. Herbst, E., and Apffelstaedt, M.: Malformations of the Jaws and Teeth, London, Oxford University Press, 1930.

natal arrest of approximately two months duration. This arrest was unusually severe and caused marked neonatal hypoplasia of enamel and dentin.

Most of the ameloblasts that were active at birth ceased to appose enamel, so that little or no postnatal enamel was formed. The formation of dentin, in contrast, continued after birth. However, only one third of the odontoblasts recovered. This resulted in a reduced number, disturbed course and atypical arborization of the dentinal tubules at the neonatal ring.

The dental tissues formed in utero were normal in structure and calcification. The postnatal dentin was poorly calcified.

The observations in this study offer additional evidence that the forming and calcifying human dental tissues are sensitive to, and constitute a permanent accurate record of, systemic disturbances.

POSTSCRIPT

The present knowledge of the interrelationship between the human dental apparatus and the rest of the body has been made possible by the active cooperation of pathologists in procuring autopsy material for microscopic study. It is hoped that additional information will result from a continued and possibly increased collaboration between hospital pathologists and dental research investigators.

COMPARISON OF CUTANEOUS LESIONS PRODUCED IN RABBITS BY INTRACUTANEOUS INOCULATION OF SPIROCHETES FROM YAWS AND SYPHILIS

HENRY W. FERRIS, M.D.

AND

THOMAS B. TURNER, M.D.

NEW YORK

In a previous paper [1] were presented the results of a comparative histologic study of representative cutaneous lesions of yaws and syphilis in man, comprising one phase of a broader investigation of the relationship of the one disease to the other. All the lesions studied occurred in persons living in Jamaica, British West Indies. From several of these same persons strains of Treponema pertenue and Spirochaeta pallida were established in rabbits and propagated in serial passage by intratesticular inoculation.[2] In order to compare further the lesions produced by strains of spirochetes from yaws and syphilis, a series of rabbits were inoculated intracutaneously with three strains of T. pertenue and a similar series with three strains of S. pallida. The reaction of the skin at each site of inoculation was noted, and at regular intervals after inoculation sections of skin were removed for histologic study. In the present paper will be presented the results of a study of this material.

EXPERIMENTAL PROCEDURE

The main purpose of these experiments was to study the histologic changes produced in the skin of a susceptible host by the inoculation of yaws or syphilis spirochetes when the reaction occurred under conditions in which the infectious agent was the only major variable factor. Thus the rabbits used in the experiments were animals of approximately the same age and were selected at random from the usual laboratory stock. During the course of the experiment all the animals were caged and fed alike. Differences in reaction between the rabbits inoculated with T. pertenue and those inoculated with S. pallida can, therefore, probably be ascribed to differences obtaining between the two types of organisms. It should be emphasized that no attempt is being made to compare the cutaneous lesions of yaws or syphilis in rabbits with the cutaneous lesions of these diseases in man.

Three strains of T. pertenue (strains YB, YC and YD) and three strains of S. pallida (strains S3, S5 and S6) were used in the experiments. The first three

From the Department of Pathology of Cornell University Medical College and the Laboratories of the International Health Division of the Rockefeller Foundation in New York.

1. Ferris, H. W., and Turner, T. B.: Arch. Path. **24**:703, 1937.
2. Turner, T. B.: Am. J. Hyg. **25**:477, 1937.

strains had been obtained from persons who presented characteristic signs of yaws, and the second three, from persons showing characteristic signs of syphilis, all living in Jamaica. At the time the present experiments were made, each strain of S. pallida had been through six serial passages in rabbits, and the strains of T. pertenue through eight, six and four serial passages, respectively. Material for inoculation was obtained by emulsifying in saline solution a portion of rabbit's testis which was the site of an active yaws or syphilis lesion. The emulsion was centrifuged to rid it of gross particles of tissue, and the supernatant fluid was used for inoculation. In each instance the inoculum contained numerous actively motile spirochetes. Each strain of spirochetes was used for inoculating two rabbits, each rabbit being inoculated intracutaneously in six areas on the back, from which the hair had been removed by clipping. The inoculums of the several strains, with the exception of strain YB, contained approximately the same number of spirochetes, as estimated by dark field examination, and 0.1 cc. of the suspension was employed as the individual dose. The suspension of strain YB of T. pertenue contained fewer spirochetes than did the other suspensions, and 0.2 cc. was used as the individual dose of this material.

In all, then, seventy-two areas were inoculated, twelve with each of the three strains of T. pertenue and the same number with each strain of S. pallida, a pair of rabbits being used for each strain, with six areas per rabbit. Animals were examined weekly or more often and notes made on the macroscopic appearance of the areas receiving the injections. In each pair of rabbits inoculated with the same strain, the inoculated areas were numbered consecutively from 1 to 12. Beginning one week after inoculation, area 1 of each pair of animals was excised for histologic study, and each week thereafter an area was removed in numerical order. Some changes in this order were made necessary because of death of one animal of a pair, but it should be noted that the inoculated areas were removed according to a prearranged schedule and not according to the size or the appearance of the lesion on the day of removal. At the time of inoculation, the point at which the suspension of spirochetes was deposited was carefully marked with dye, and on removal of the area its center, or the center of the lesion when one was present, was again marked in order that microscopic sections from this portion of the area could be identified.

Tissue specimens were fixed in neutral solution of formaldehyde U. S. P. diluted 1:10, embedded in paraffin and cut in the usual manner. Sections were stained with hematoxylin and eosin, and, for the purpose of demonstrating spirochetes, by the Warthin-Starry method of single section silver impregnation.[3] The histologic technic, including the silver impregnation, was carried out by Miss Frances Fraser.

RESULTS

The comparison of the results of the inoculations in the two groups of rabbits was made on the basis of the gross and microscopic changes at the sites of inoculation and the distribution of spirochetes in the lesions. In spite of the fact that the inoculums for all the strains of T. pertenue and S. pallida seemed to contain approximately the same number of organisms, there was a pronounced difference in the evolution of the lesions of yaws as compared with those of syphilis. This

3. Farrier, R., and Warthin, A. S.: Am. J. Syph. **14**:394, 1930.

difference was uniform for each of the three strains of T. pertenue and S. pallida. One week after inoculation, in both the animals with syphilis and those with yaws, there was slight redness in nearly all the inoculated areas. By the end of the second week, however, most of the syphilitic lesions had increased in size to form papules approximately from 2 to 5 mm. in diameter. A few of the lesions of yaws had increased in size, but a number of others had subsided to such an extent that no macroscopic lesion was visible. Four weeks after inoculation, all the remaining areas in the rabbits with yaws were normal to macroscopic examination, while a majority of the syphilitic lesions had become larger and resembled typical hunterian chancres. This difference in the two groups of rabbits was even more pronounced after six weeks. The number of inoculated areas showing slight lesions, well developed

Number of Areas Inoculated with T. pertenue or S. pallida Showing no Lesions, Slight Lesions or Well Developed Lesions at Various Intervals After Inoculation

Days After Inoculation	Yaws Rabbits				Syphilis Rabbits			
	Number of Areas Unexcised	Number of Areas Showing			Number of Areas Unexcised	Number of Areas Showing		
		No Lesion	Slight Lesion	Well Developed Lesion		No Lesion	Slight Lesion	Well Developed Lesion
7	36	0	36	0	36	6	30	0
14	28	10	16	2	33	0	33	0
21	25	7	18	0	30	1	25	4
28	21	21	0	0	27	1	7	19
42	13	13	0	0	20	0	2	18

lesions or no lesions at various periods after inoculation are given in the table. The two yaws lesions present after two weeks and classified as "well developed" were not nearly so large as a majority of the syphilitic lesions similarly classified. These yaws lesions were not elevated and showed little or no induration. They were classified as "well developed" largely because of superficial ulceration of the surface of the lesion.

Histologic Changes.—As may be seen in figure 1, there was a conspicuous difference in the intensity of the skin reactions induced by T. pallidum and S. pallida.

The inflammatory reaction in the early (one week) cutaneous lesions of syphilis (fig. 2 *A*) consisted in an infiltration, chiefly by lymphocytes, with a few larger mononuclear cells and a scant number of plasma cells and polymorphonuclears, of areas about hair follicles and small blood vessels in the superficial part of the dermis. Endothelial cells of capillaries were swollen, and there was proliferation of fibroblasts. This was followed by more diffuse distribution of the cells and occurrence of

deeper perivascular accumulations. The early lesions of yaws were similar but usually smaller (fig. 3 *A*). In both syphilis and yaws the epidermis over the lesion was slightly thickened, owing to the formation of one or two extra layers of epithelial cells.

More advanced lesions of syphilis presented varying degrees of necrosis of the epidermis and underlying dermis, with abundant exudation of polymorphonuclear leukocytes (fig. 2 *B*). The deeper tissues were edematous and contained polymorphonuclears and lymphocytes in varying numbers, with a few plasma cells. Proliferation of fibroblasts

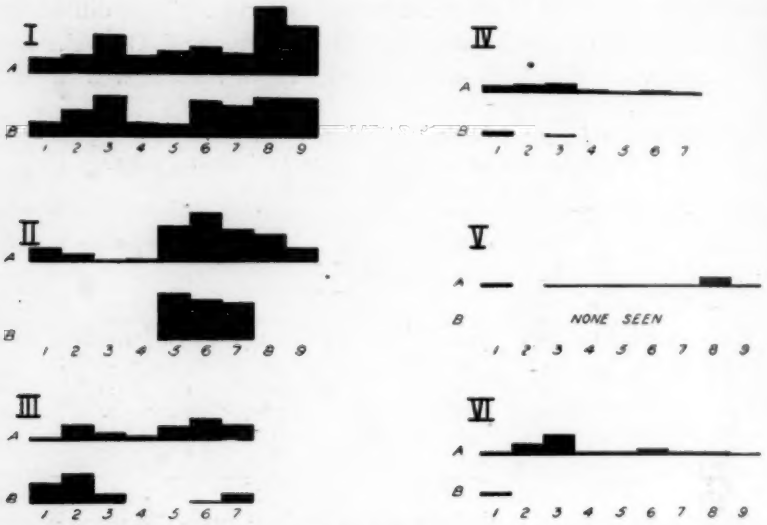


Fig. 1.—Graphic representation (based on microscopic study) of the degree of inflammatory reaction (*A*), and the number of spirochetes (*B*) in the cutaneous lesions produced in rabbits by intracutaneous inoculation of three strains of S. pallida (left) and three strains of T. pertenue (right) as follows: I, syphilis strain S3; II, syphilis strain S5; III, syphilis strain S6; IV, yaws strain YB; V, yaws strain YC; VI, yaws strain YD. The arabic numbers refer to the weeks after inoculation.

produced an increase of young connective tissue, which was often edematous and infiltrated by many cells. The perivascular infiltrations were seen chiefly at the margins of the lesion and in the deeper parts. Even the subcutaneous muscle became involved by the exudation and connective tissue proliferation. Beneath the crust, epithelium regenerated to form a layer considerably thicker than normal, and after the drying and falling off of the crust, this layer covered the somewhat depressed surface of the lesion. Completely healed lesions were not represented in the series.

The maximal lesion in the yaws series studied microscopically, seen at three weeks in an area receiving strain YD, was characterized by edema and the presence of a few polymorphonuclear leukocytes in the

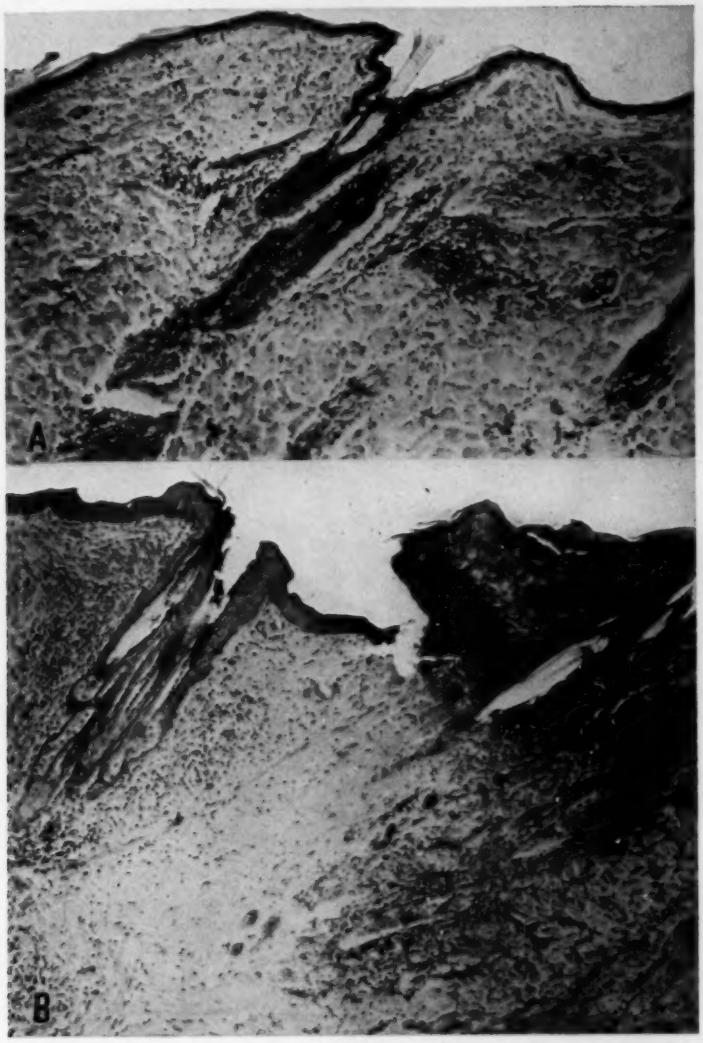


Fig. 2 (rabbit 207).—*A*, section of skin taken one week after intracutaneous injection of S. pallida (strain S3), showing an infiltrate, predominantly of lymphocytes, about hair follicles and small blood vessels of the dermis (see fig. 4 *A*); *B*, section taken three weeks after intracutaneous injection of S. pallida (strain S3), showing a perifollicular infiltrate composed chiefly of lymphocytes. The epidermis is thickened. On the right is superficial necrosis, with an underlying exudate of polymorphonuclear leukocytes. (In this area spirochetes are especially numerous in silver impregnations.) Both *A* and *B* were prepared with hematoxylin and eosin; magnification, × 81.

dermis, as well as by perivascular infiltrates of lymphocytes. In general, however, the yaws lesions, less developed at all stages, after this period became inconspicuous microscopically as well as grossly, consisting essentially in scant superficial lymphocytic infiltration.

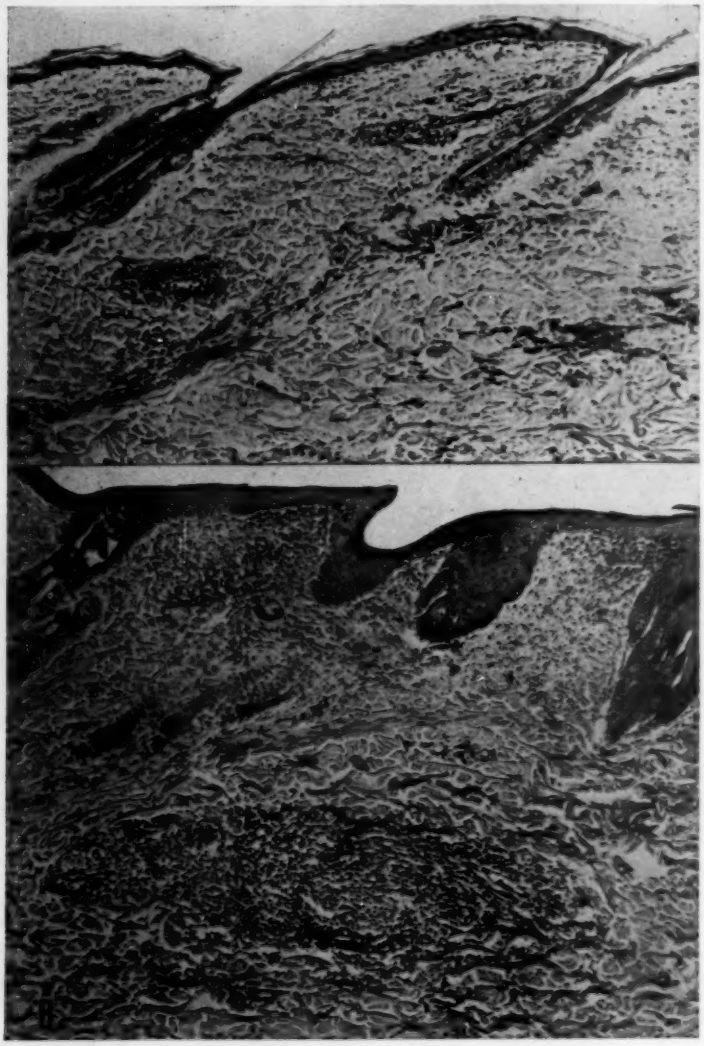


Fig. 3.—*A*, (rabbit 213), section of skin taken one week after intracutaneous injection of T. pertenue (strain YB), showing a scant, scattered perivascular and perifollicular infiltrate composed predominantly of lymphocytes (see fig. 4 *B*); *B* (rabbit 214), section taken three weeks after intracutaneous injection of T. pertenue (strain YB), showing more extensive and deeper cellular infiltration. By silver impregnation, one spirochete was found in the surface epithelium, and one in a deep focus of lymphocytes. Both *A* and *B* were prepared with hematoxylin and eosin; magnification, × 81.

It is noteworthy that polymorphonuclear leukocytes were seen in considerable numbers only in lesions which had undergone ulceration, and hence they were relatively inconspicuous in the lesions of yaws.

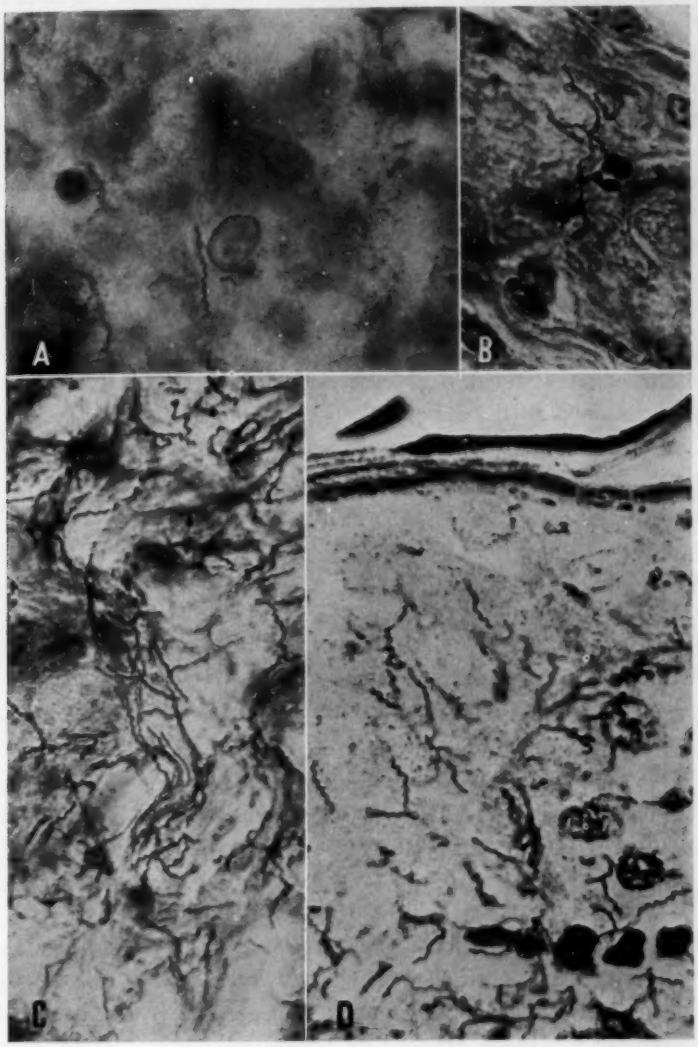


Fig. 4.—*A* (rabbit 207), section of skin taken one week after injection of *S.* pallida (strain S3), showing spirochetes in a hair follicle (see fig. 2 *A*); *B* (rabbit 213), section taken one week after injection of T. pertenue (strain YB), showing spirochetes in a hair follicle (see fig. 3 *A*); *C* (rabbit 210), section taken seven weeks after injection of S. pallida (strain S5), showing numerous spirochetes in a deep edematous area of the dermis; *D* (rabbit 208), section taken nine weeks after injection of S. pallida (strain S3), showing spirochetes in the epidermis and dermis near the edge of the ulcer. The staining technic for all sections was the Warthin-Starry silver impregnation; magnification, × 1,500.

Spirochetes.—Of the lesions produced by each species of organism, the earliest specimen was removed one week after inoculation. At this stage spirochetes were seen in two of the three syphilitic lesions. They were numerous within the epithelium of several hair follicles about which cellular infiltration was conspicuous (fig. 4 *A*) and were also seen in smaller number in the adjacent epidermis. A similar relation of spirochetes to the walls of small blood vessels with perivascular infiltration was observable. In two of the one week lesions of yaws, organisms were also seen, in smaller numbers, limited to the hair follicles and epidermis (fig. 4 *B*). With the exception of the three week lesion of strain YB, in which only one spirochete was seen in the epidermis and one in a fairly deep focus of lymphocytes, T. pertenue was not demonstrated in any further lesions of yaws.

In the later lesions of the syphilitic series, spirochetes were increased in number (fig. 4 *C*), being found in the greatest quantities in the areas of inflammation and early fibrosis below the necrotic crust, in some instances as deep as the subcutaneous muscle. In several lesions they were in vast numbers, forming grayish streaks that were visible with low magnification in silver impregnations. Organisms were not seen in the eight and nine week lesions due to strain S5, which had undergone much regression, with fibrosis and diminution of the inflammatory reaction.

It is of interest that with strain S3 of S. pallida several small cutaneous lesions were present a few millimeters from the chancres, as satellites. One was excised with an eight week lesion and two with a nine week chancre. Histologically they were much the same in appearance as the primary sores. Noteworthy is the observation that there was no direct continuity, grossly or microscopically, between the primary lesions and the satellites, and that while spirochetes were numerous in the satellites no organisms were demonstrable in the intervening tissue.

COMMENT

It is evident that each of the three strains of S. pallida used in these experiments produced much larger lesions than did any of the three strains of T. pertenue. Since all these strains had been through approximately the same number of passages in rabbits, and since the inoculums of the several strains contained about the same number of spirochetes, and the animal hosts and the environmental influences to which these hosts were subjected were similar, it is probable that the differences in reaction to the two types of infection can be attributed to inherent differences between S. pallida and T. pertenue. This difference, consistent with that seen in rabbits following intratesticular inocu-

lation,[4] may be, of course, in virulence alone, but if so, it is one shared by each of the three strains of the two organisms.

The importance of an increase of large mononuclear cells (macrophages) in the resolution of syphilitic lesions in the rabbit has been stressed by a number of investigators.[5] These observed that an artificially produced increase of macrophages was associated with increased rapidity of resolution. They did not, however, utilize silver impregnation to demonstrate the relation of the phagocytes to the spirochetes. It is noteworthy that while a few groups of large mononuclear cells were found in the healing syphilitic lesions which we studied, no evidence of phagocytosis of organisms by them was seen, and cells of this type were not abundant in the other lesions of either syphilis or yaws. Nor was there evidence of ingestion of spirochetes by any other cells of the fixed tissue or of the exudate in the lesions of either yaws or syphilis. This is in agreement with observations made in the study of cutaneous lesions of yaws and syphilis in human beings [1] but contrary to the observation of Levaditi and his associates,[6] who described the presence of typical spirochetes or atypical coiled or granular forms in lymphocytes and plasma cells in syphilitic scrotal lesions of rabbits. On the other hand, studies by Beck [7] failed to reveal phagocytosis of the spirochetes of syphilis in vitro.

Coiled, granular or poorly stained forms of S. pallida were numerous in some of the cutaneous lesions of rabbits in this series. They were usually situated in areas of necrosis or of extensive leukocytic exudation. The authors consider these forms as representing actual degeneration, rather than involution, of the organisms, as suggested by Levaditi.

The frequent occurrence in advanced lesions of large numbers of spirochetes in bundles of connective tissue, lying between the fibrils and parallel to them, is worthy of note. This intimate relation indicates that the micro-organisms can proliferate within collagen bundles.

From the standpoint of pathogenesis it is of interest to consider the distribution of the organisms at different periods of development of the lesions. We have not yet studied the disposition of the spirochetes immediately after their injection and up to a week later. In the earliest

4. Turner, T. B., and Chesney, A. M.: Bull. Johns Hopkins Hosp. **54**:174, 1934.

5. Morgan, H. J.: Tr. A. Am. Physicians **45**:67, 1930. Morgan, H. J.; Harris, S.; Cunningham, R. S., and Tompkins, E. H.: J. Clin. Investigation **10**:661, 1931. Cunningham, R. S.; Morgan, H. J.; Tompkins, E. H., and Harris, S.: Am. J. Syph. **17**:515, 1933. Harris, S., Jr.; Tompkins, E. H.; Morgan, H. J., and Cunningham, R. S.: ibid. **18**:333, 1934.

6. Levaditi, C.; Vaisman, A.; Shoen, R., and Manin, Y.: Ann. Inst. Pasteur **56**:251, 1936.

7. Beck, A.: J. Path. & Bact. **44**:399, 1937.

lesions of both syphilis and yaws excised (one week after inoculation) the spirochetes were located in the epidermis and related structures, the hair foliicles. In the syphilitic lesions the organisms were also seen in the adjacent dermis in and about small blood vessels. The multiplication of T. pertenue appeared to be arrested early, and the inflammatory reaction produced was slight. S. pallida, however, continued to multiply in both epidermis and dermis, spreading deep into the tissues, accompanied by an infiltrate of lymphocytes and other cells. In the healing lesions the organisms disappeared before the inflammation completely subsided.

SUMMARY

In this series of rabbits, the initial lesion produced by T. pertenue following intracutaneous inoculation was less rapid in its development and did not attain the size of the lesion produced by S. pallida.

In lesions of comparable size, the histologic changes are qualitatively the same in yaws and syphilis, consisting predominantly in lymphocytic infiltration about hair follicles and small blood vessels. The lesions of yaws subside rapidly, while those of syphilis become indurated by new formation of fibrous tissue.

The distribution of the spirochetes in hair follicles and epidermis is essentially the same in comparable lesions of the two diseases.

In the larger lesions of syphilis, necrosis and an exudation of polymorphonuclear leukocytes are prominent, and spirochetes are numerous. In the lesions of yaws there are few spirochetes, and they disappear rapidly.

There is no evidence of phagocytosis of spirochetes by any type of cell in either yaws or syphilis.

NEW AND SIMPLE METHOD FOR THE DETECTION OF BLOOD BY HEATING

A. M. MOODY, M.D.
SAN FRANCISCO

FREDERICK PROESCHER, M.D.
SAN JOSE, CALIF.
AND
JESSE L. CARR, M.D.
SAN FRANCISCO

Because of the medicolegal importance of the identification of blood and the difficulty in eliminating conflicting substances, new methods are constantly being suggested. When the blood to be identified is contaminated with organic or inorganic substances, decomposed by atmospheric, chemical or physical agencies, exposed to high temperature with complete carbonization or simply reduced to its inorganic constituents, either positive reaction with contaminating substances or in the case of heating a total absence of reaction with any of the presumptive tests has resulted in confusion and often inconclusive evidence. Leading textbooks on forensic medicine advise spectroscopic or fluorescence tests on presumptive blood stains when there is a conflicting substance or in the absence of reaction with one of the commoner peroxidase tests. If very small quantities of blood are present, however, the resulting dark solutions are difficult to analyze, and in the majority of cases a definite answer as to the presence or absence of blood cannot be given.

Literature on the physical changes of blood is limited to information drawn from tests made on blood at the normal temperature of the body or room. The physical behavior of blood heated beyond this point has, to our knowledge, not been carefully studied up to this time. Recently it became necessary as a matter of evidence to determine whether or not blood stains could be identified after exposure to temperatures above normal ranges, but no references were available on which one could base accurate conclusions. Smith and Glaister [1] had reported the loss of the precipitin test for blood when the blood was heated above 392 F. (200 C.) for five minutes. Hurd [2] had reported

---

From the St. Francis Hospital.

1. Smith, S., and Glaister, J., Jr.: Recent Advances in Forensic Medicine, Philadelphia, P. Blakiston's Son & Co., 1931.

2. Hurd, C. D.: The Pyrolysis of Carbon Compounds, New York, The Chemical Catalog Company, Inc., 1929.

studies on the pyrolysis of numerous carbon compounds and their physical behavior at high temperature but had not included any specific studies on blood. Outside of these findings and a few incomplete studies in German literature, no observations of blood heated above 320 F. had been made. Scott [3] had studied the structure of individual cells by microincineration but had not utilized his method for medicolegal identification of blood. The necessity of a satisfactory and dependable method of identifying grossly contaminated, carbonized or completely incinerated blood became obvious.

To determine whether or not blood could be reliably identified as such after incineration, a large series of experiments were made wherein blood samples as such or mixed with organic or inorganic material were heated from 212 F. (100 C.) to and including 1,600 F. (871 C.). The incinerated samples were studied as to gross and microscopic appearance, for chemical reaction to benzidine and leukomalachite green, for formation of hematin and hemochromogen crystals and for formation of hematoporphyrin.

METHOD OF HEATING

The incineration experiments were made in large gas furnaces with observation windows, as used for metallurgic work, with a small electrically heated incinerator, and in a solder bath, each with exact temperature controls. For the tests approximately 5 cc. amounts of clotted whole blood were placed in small steel and iron cups (1¼ inches [3 cm.] in diameter by ⅝ inch [1.6 cm.] in height) and heated in a drying oven at 320 F. (160 C.) for three hours. Iron cups were used because a reliable test should eliminate all reactions by metallic iron. For further incineration they were placed in the high temperature gas furnaces. In view of the fact that the drying temperature was 320 F. (160 C.), the incineration temperatures in the gas furnaces were begun at 500 F. (260 C.) and carried to 1,600 F. (871 C.) (fig. 1). The procedure of incineration was as follows: All samples were placed in the furnace at the same time, and the temperature was equalized; when the temperature reached a stable level of 440 F. (227 C.), it was run up to 500 F. (260 C.) where it was held for five minutes, at which point a sample was removed. The temperature was then raised another 100 F. (38 C.) and held for five minutes, and then another sample was removed. This method was followed until the last sample was removed at 1,600 F. (871 C.) after being held at this temperature for five minutes. The time from one 100 F. rise to another was approximately thirty minutes. On removal, each sample was carefully scraped from the cup, placed in a specially prepared glass container, marked, corked and taken to the laboratory for further study.

OBSERVATIONS WITH LARGE SAMPLES OF BLOOD

Knowing nothing of the physical behavior of blood beyond 500 F. (260 C.), those observing these samples of blood were surprised to note

3. Scott, G. H.: Protoplasma **20**:133, 1933.

the distinctive and characteristic changes in the samples. It gradually became evident that not only could blood be identified after heating to 500 F. (260 C.) and over but that heating blood to such high temperatures actually offered one of the best means of identifying blood. The physical changes were so characteristic that not only could the blood be identified but the approximate temperature to which it had been heated was physically evident. Dried blood melts at a little above 500 F. (260 C.) and boils up to a glairy-like bubble with a shiny, refractile surface. This melting point, observed in the iron cups, has been checked by heating blood drawn into fine glass capillaries, the blood melting between 500 and 515 F. At 600 F. (315 C.) the escape of gases in the melted, fluid blood causes a further bubbling and eruption of the blood mass up to and over the brim of the cup, offering a glazed and shiny black cone or sphere of material in which there are many tiny vacuoles. At 700 F. (371 C.) much of the gas has escaped, and the blood mass begins to shrink back down in the cup, forming a black mass, which grossly

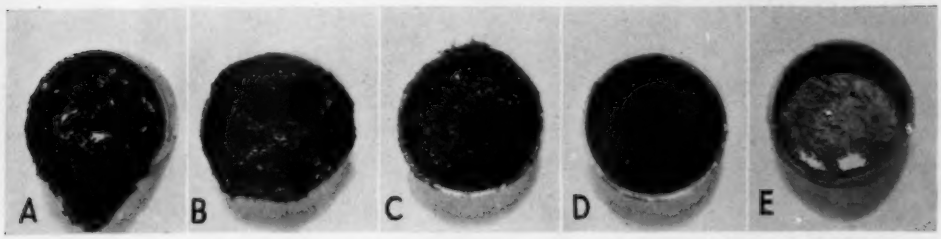


Fig. 1.—Blood in iron cups heated to various temperatures: *A*, 600 F.; *B*, 700 F.; *C*, 800 F.; *D*, 900 F.; *E*, 1,000F.

appears to be formed by innumerable fine crystals. At 800 F. (426 C.) the blood mass is still more finely divided, is reduced in volume and has a fine mossy appearance, as though it were existing in a physical form composed of many tiny branches or rods. At 900 F. (482 C.) it is reduced to a very fine webby dull black mass, and at 1,000 F. (537.8 C.) it has assumed the appearance of finely granular red powder. This appearance is maintained above the point of 1,000 F., gradually becoming finer in texture up to 1,200 F. (649 C.), where it apparently reaches a final stage.

Because of these odd physical changes, samples were placed under a dissecting microscope and examined by reflected light on a white background. By such examination the physical changes are even more striking. At 500 F. (260 C.) the blood masses reveal large shiny fields with an enamel-like surface. At 600 F. (315 C.) the masses are very black again and refractile, and frequent vacuoles can be seen within the mass

(fig. 2). At 700 F. (371 C.) a definite pattern similar to a multiple collection of pentagonal rings can be seen, which suggest rings structurally of the pyrrole type (fig. 2). These have relatively fixed black sides and are joined to all the others, while the spaces between the sides are filled with a thin, brittle, translucent and slightly brownish yellow membrane, resembling a thin layer of dry pyroxylin. They appear like a number of compressed springs being slowly released but having straight and irregularly angular sides instead of circular coils. At 800 F. (426 C.) the walls become thinner and appear somewhat curved, and

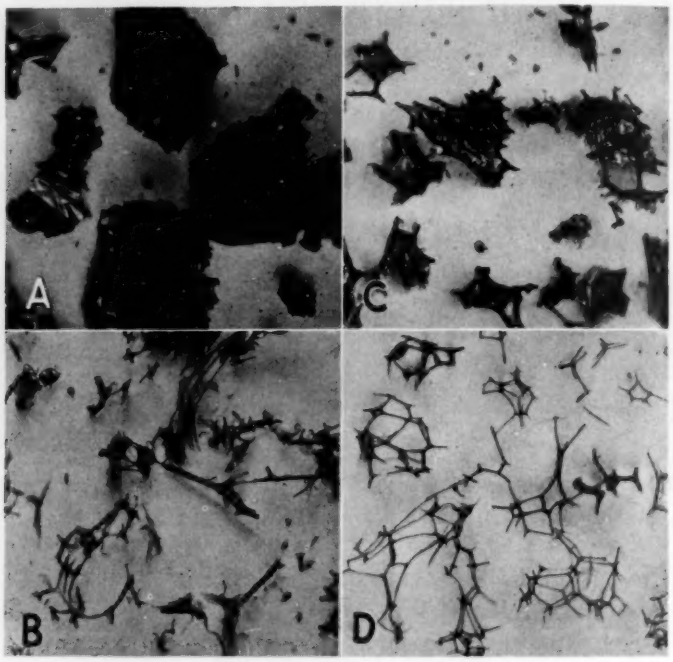


Fig. 2.—Physical changes in whole blood at various temperatures: *A*, 600 F.; *B*, 700 F.; *C*, 800 F.; *D*, 900 F.

the central areas begin to disappear, while in their places small fluffy red masses are seen (fig. 2). Scattered throughout, some fine membranes can still be seen between the sides or bars of the rings, but these side structures are becoming more netlike and fragile. At 900 F. (482 C.) the samples are grayish black and in the dissecting microscope assume the optimal development of a definite structural form. The blood at this stage is a soft, fragile combination of four-sided and five-sided figures united in an irregular but definite manner, and forming a huge, irregular and apparently purposeless scaffolding, formed of fine black bars. The structure is quite complicated, and when viewed stereo-

scopically, it gives one an impression of resemblance to the wire models used in teaching molecular structure and rings (fig. 2). Whether there is any relationship or not between the chemical structure of the blood constituents (heme, globulin and inorganic substances) is not yet determined, but this possibility is an interesting speculation in view of the consistency of development of these forms. At 1,000 F. (537.8 C.) the bulk of the mass, which has decreased to at least one fiftieth of its maximal developed volume, is grossly a rusty red fluffy dust, but in the binocular microscope a few of these pentagonal forms are still present, though delicate and breaking up (fig. 3). At 1,100 F. (593 C.) none of these structures remain, and only an amorphous red dust is left. A few black bars can be defined, but the structure is now eliminated entirely by the heat (fig. 3).

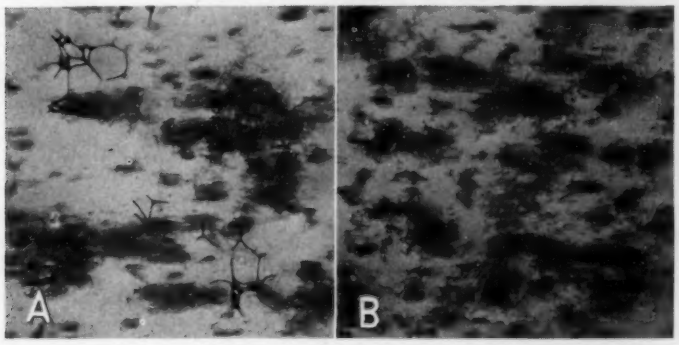


Fig. 3.—Physical appearance of blood heated at temperatures over 900 F.: *A*, 1,000 F.; *B*, 1,100 F.

Not realizing at the time of the initial experiments that this was a specific change for blood, physically speaking, a number of other substances were similarly heated and studied in the dissecting microscope. Among these were human, guinea pig, rabbit, chicken and sheep blood and serum. Chlorophyll, egg white, egg yolk, various mixtures of vegetables, grasses and other organic substances were also used, both alone and mixed with blood. In every instance in which the incinerated mass contained blood, either pure or in mixtures, the characteristic structures developed. Some other incinerated organic substances of large molecular structures showed characteristic heat structures, but they could be readily distinguished from those of blood. After very little experience in examining these specimens, it became possible not only to identify blood alone but to identify it when it was mixed with innumerable other materials and to exclude blood from a mixture of these materials if the ringlike structures were not found.

OBSERVATIONS WITH SMALL SAMPLES OF BLOOD

Having demonstrated the specific structural changes which take place by incinerating large quantities of pure and contaminated blood, experiments were made to determine the applicability of the incineration method for small quantities of blood.

For these experiments a small, electrically heated laboratory furnace was used, with which a maximal temperature of nearly 1,832 F. (1,000 C.) could be obtained. The temperature was controlled with a standardized thermocouple and an adjustable rheostat and checked with a nitrogen thermometer.

Human blood as well as blood from a number of vertebrate animals (horse, beef, rabbit, guinea pig, sheep, cat, rat, pig and chicken) was first dried in a vacuum desiccator over phosphoric anhydride; when thoroughly dried, it was finely ground and incinerated in silica or platinum dishes. Quantities of from as small as 1 mg. up to 0.1 and 0.25 Gm. were used. The incineration temperatures began at 392 F. (200 C.) and were carried up to, and including, 1,552 F. (900 C.) with intervals of from five to sixty minutes.

In each of these samples the characteristic ring structures developed when the temperature of 700 F. was reached and, while in smaller number in the minute samples, the structures were as characteristically reliable as those that developed when larger volumes of blood were burned.

SUMMARY

Study of the physical changes which occur when blood and a few other organic substances are heated to high temperatures has demonstrated that dried blood melts at from 500 to 515 F. (260 to 268 C.), that blood after further heating above 700 F. assumes a specific structure, that this structure is easily identified in quantities as small as 1 mg. even when the blood has been mixed with inorganic or other organic substances and that the physical changes are specific not only for blood but also for the degree of heat used.

St. Francis Hospital, San Francisco (Dr. Moody).
Santa Clara County Hospital, San Jose (Dr. Proescher).
University of California Medical School, San Francisco (Dr. Carr).

PATHOLOGIC ANATOMY OF HYPOPHYSIS AND ADRENALS IN ANENCEPHALY

D. MURRAY ANGEVINE, M.D.
NEW YORK

Many anatomists and pathologists have written of the hypophysis and adrenals in anencephaly. Meckel [1] (1812) stated that the adrenal is small or lacking in anencephaly, and he did not find it in 2 of 9 cases examined. Ballantyne [2] found that in 45 cases the pituitary was rarely present and that the adrenals were usually absent or defective. Browne [3] was unable to find the hypophysis in 4 cases and considered that "apituitarism" was the cause of anencephaly. He reported absence of the adrenals. Mauksch [4] examined 9 cases and found the hypophysis in every instance, but all three lobes were present in only 2 cases. The adrenals were not mentioned. Barlow [5] reported that he was able to find the hypophysis in 4 instances, and in only a single instance was a pars nervosa present. He described the adrenals as normal. Kohn [6] found the pars anterior of the hypophysis in all of 11 cases examined. The pars intermedia was usually absent, and the pars nervosa was present in only 2 instances. Kiyono [7] was able to find the hypophysis in 7 of 11 cases, and the posterior lobe was found in 3 of these. Ettinger and Miller [8] examined 9 specimens and found the hypophysis in 8, of which 3 had a pars nervosa. The adrenal was present in only 2 cases. Covell [9] made a detailed study of the hypophysis in 32 anencephalic monsters and found it present in every one. Of 17 of the specimens studied in detail, 6 had all three lobes present; 11 had no pars nervosa, and of these only 4 had a pars intermedia, the gland in the rest being composed of a pars anterior alone. Vaclav,[10] in a careful study of a large series of

From the Department of Pathology, Cornell University Medical College.

1. Meckel, J. F.: Handbuch der pathologischen Anatomie, Leipzig, C. H. Reclam, 1812.

2. Ballantyne, J. W.: Antenatal Pathology and Hygiene, New York, William Wood & Company, 1902.

3. Browne, F. J.: Edinburgh M. J. **25**:296, 1920.

4. Mauksch, H.: Anat. Anz. **54**:248, 1921.

5. Barlow, D. L.: Brit. M. J. **1**:15, 1923.

6. Kohn, A.: Arch. f. mikr. Anat. **102**:113, 1924.

7. Kiyono, H.: Virchows Arch. f path. Anat. **257**:441, 1925.

8. Ettinger and Miller: Tr. Roy. Soc. Canada **5**:247, 1926.

9. Covell, W. P.: Am. J. Path. **3**:17, 1927.

10. Vaclav, J.: Sborn. lék. **28**:399, 1927.

anencephalic monsters, found the adrenals in all of 56. In an additional case only a single adrenal was present, and this was associated with absence of a kidney. He stated that he found the hypophysis constantly. A detailed histologic study was made on 30 specimens.

From the last two large series it seems that both the hypophysis and adrenals are present in every anencephalic monster and that their reported presence or absence depends on the diligence and care with which they are searched for. Malpas [11] stated that the pituitary was normal in many anencephalic monsters; however, in a description of 9 cases of anencephaly, he reported it to be absent in 3 and not looked for in the others. He described the adrenals as normal in 2, small in 1 and absent in 6 cases. In a recent case reported by Broder [12] the hypophysis was not found.

The following study was undertaken to determine whether the hypophysis and adrenals are always present in anencephaly, and to determine, if possible, how lesions in the two glands are related. It also seemed desirable to determine whether the adult type of adrenal found in anencephaly is the result of lack of development from the beginning or of subsequent prenatal atrophy.

The other glands of internal secretion were carefully studied in most instances, and special attention was given to the presence of other congenital anomalies.

An attempt was also made to correlate some of the anatomic changes with clinical signs observed during pregnancy and especially with hydramnion.

CLINICAL HISTORIES

A history was available for, and a complete autopsy was performed on, each of 15 anencephalic monsters. Five additional specimens were obtained on which either no histories were available or the necropsies were limited. Each specimen is designated by the same number in the accompanying table and in figures 2 and 4.

The ages of the mothers ranged from 22 to 42 years; the average age was 28.8 years. Seven of the 15 monsters resulted from first pregnancies. Four of the mothers had had from one to four miscarriages. One had had premature labor in pregnancies prior to the birth of the anencephalic monster. The obstetric histories of the others were not significant in this relation.

A definite diagnosis of hydramnion was made in 8 of the 15 pregnancies. In case 8 the diagnosis was not made before delivery. The diagnosis was uncertain in 2 other cases, but there probably was

11. Malpas, P.: J. Obst. & Gynaec. Brit. Emp. **49**:1046, 1933.

12. Broder, S. B.: Am. J. Path. **11**:761, 1935.

hydramnion because in both cases the large size of the abdomen was noted and in case 10 lightening did not occur. The obstetrician noted that the amount of fluid was abundant in case 16, but a diagnosis of hydramnion was not recorded. No roentgen examination was made in these cases. Roentgenograms were taken in 7 cases in which a diagnosis of hydramnion was made before delivery, and each showed the presence of an anencephalic monster in utero. A roentgenogram was made in case 17 because the pregnancy had gone forty-two days beyond term.

Of 12 cases in which the duration of the pregnancy was recorded, in 6 the delivery was regarded as premature, in 6 as at term and in 3 as

Twenty Cases of Anencephaly

Case	Weight of Anencephalic Monster, Gm.	Sex	Hypophysis*			Weight of Adrenals,* Gm.	Choroid Plexus*	Hydramnion†	Roentgen Diagnosis
			Pars Anterior	Pars Intermedia	Pars Nervosa				
1	1,030	F	+	0	+	+2.5	0	+	+
2	1,100	F	+	+	0	+1.0	+	0	—
3	1,140	F	+	0	0	+1.0	0	0	—
4	1,170	M	+	0	0	+1.0	+	No history available	—
5	1,360	F	+	0	0	+ —	—	+	+
6	1,580	M	+	+	+	+6.0	+	+	+
7	1,600	F	+	0	0	+1.9	—	+	+
8	1,700	F	+	+	+	+0.3#	+	+	—
9	2,000	M	+	0	0	+ —	+	+	+
10	2,000	F	+	0	0	+1.2	+	Large abdomen	—
11	2,000	M	+	+	0	+ —	—	No history available	—
12	2,060	F	+	0	0	+0.3	0	0	—
13	2,300	F	+	+	0	+0.2	0	0	—
14	2,350	F	+	0	0	+0.4	+	+	+
15	2,400	M	+	0	0	+0.2	+	+	+
16	2,650	F	+	0	+	+0.6	+	Abundant	—
17	3,100	F	+	+	+	+0.6	—	0	+
18	2,960	F	+	0	0	+0.8	—	No history available	—
19	.....	F	+	0	0				
20	.....	..	+	0	0				

* A plus means that the part was found; a zero, that it was not found; a dash, that it was not looked for.
† A plus means that the condition was observed, a zero that it was not present.
# Only the right adrenal was weighed.

postmature by seven, thirteen and forty-two days, respectively. The weights varied from 1,030 to 2,960 Gm.: about half of the monsters weighed more and half less than 2,000 Gm.

ANENCEPHALIC MONSTERS

There were 13 females and 6 males in the series; the sex was not recorded in case 20. The bodies varied in length from 33 to 50 cm., and the curves of length and weight were closely parallel. Any slight differences in the ratio of height to weight were probably due to the difficulties of measuring caused by variations in shape of the malformed heads. The extremities were carefully measured in most instances, and the arms were found to be constantly longer than the legs. This observa-

tion was made by Nanagas,[13] who made detailed measurements of 57 monsters. In practically every case the subcutaneous pannus of fat was unusually thick, especially over the shoulders and thorax.

HYPOPHYSIS

It was usually impossible to recognize the hypophysis as a gross anatomic structure, but its position could usually be determined by first locating the poorly formed sella turcica. Lateral roentgenograms of the skull were made in several instances, but they gave little aid. The

Fig. 1.—Dorsal view of an anencephalic monster. The rectangular area indicated was removed for section. The letters *a a* denote sites of hemorrhagic tissue removed for section.

entire specimen was fixed in a 1:10 dilution of solution of formaldehyde U. S. P. or in Kaiserling solution, and after forty-eight hours a rectangular block of bone (fig. 1) including the sella turcica was removed. The block measured about 1 cm. transversely and 4 cm. anteroposteriorly; after decalcification it was sectioned vertically through the central part. In many instances one was then able to recognize the hypophysis as a small yellowish brown oval structure that varied greatly in outline and size. Microscopic sections of the gland

13. Nanagas, J. C.: Am. J. Anat. **35**:455, 1925.

were prepared at several levels, and those through the largest part of the gland were used in comparing one gland with another. Several specimens had been preserved in fixative for a considerable time, so that the tissue was not suitable for detailed cytologic study.

The slides were placed on an Eastman autofocus enlarger, and direct tracings were made. The various shapes of the hypophyses of 3 normal

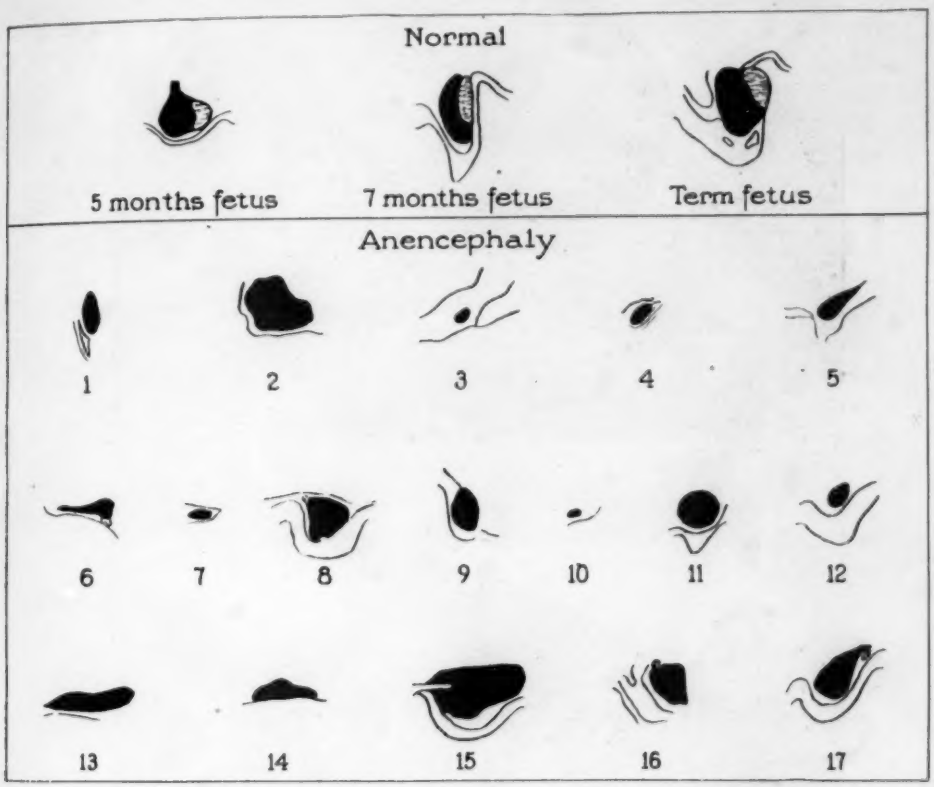


Fig. 2.—Drawings of sections of the hypophyses from 3 normal and 17 anencephalic fetuses. The anterior lobe is in solid black, and the posterior lobe is shaded. The glands are enlarged to about twice the normal size.

fetuses and 17 anencephalic monsters are shown in figure 2. In anencephaly the gland was usually composed entirely of anterior lobe, and this was present in all of the 20 cases examined (table). No relationship between the size of the gland and the size of the fetus was noted, although in general the larger fetuses had the larger hypophyses, and the shape of the glandular tissue corresponded to that of the poorly formed sella turcica. A section of the hypophysis through the sella turcica of a normal fetus that weighed 1,300 Gm. is shown in figure 3 *A*.

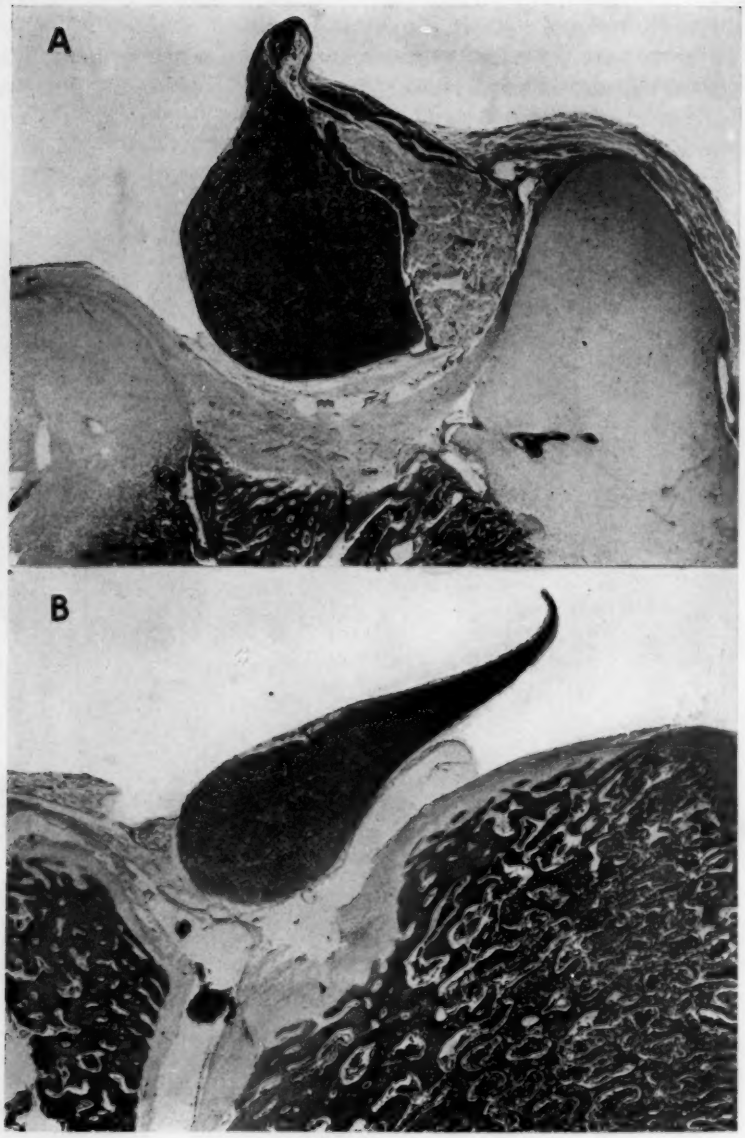


Fig. 3.—*A*, midsagittal section through the hypophysis of a normal fetus weighing 1,300 Gm.; × 13. *B*, midsagittal section through the hypophysis of an anencephalic monster (case 5) weighing 1,300 Gm.; × 13.

The hypophysis from a monster (case 5) weighing 1,300 Gm. is seen in figure 3 *B*. The sella turcica is poorly formed, and no posterior lobe is present. In a few instances there was so little hypophysial tissue that serial sections of the block were required to locate it. In case 3 the only trace of the gland was a small bit of hypophysial tissue in the pharyngeal region. There was a small pharyngeal pituitary in case 11 as well as a large anterior lobe in the usual position. A structure corresponding to the pars intermedia was recognized in 6 instances (table), and a small pars nervosa was found in 5. The three parts of the gland were found in only 3 of the 20 anencephalic specimens examined. The nerve tissue that was present varied from a few nerve fibers to a fairly well formed cerebrum with a cerebellum attached. There was no apparent relationship between the amount of nerve tissue present and the presence or absence of the pars nervosa.

The hypophyses were compared with those of normal fetuses of about the same weights, and no conspicuous differences were noted. About one third of the hypophyses were highly vascular, one third moderately vascular and the remainder only slightly so. Eosinophilic cells were present in moderate numbers in 6 cases. It is possible that they would have been observed more often had the tissue been better preserved.

ADRENALS

The adrenals were uniformly smaller than those of normal fetuses (fig. 4). They were found in all of 18 cases examined (table) but with considerable difficulty in a few instances. The right adrenal only was located by the prosector in case 8.

In figure 5 *A* is shown a transverse section through the largest part of the adrenal of a normal 1,700 Gm. fetus; the cortex is poorly differentiated, and little medulla is present. The adrenal from an anencephalic monster (case 9), 2,000 Gm. in weight, is shown in figure 5 *B*. It is much smaller than that shown in *A*. The cortex is well differentiated, and there is considerable medullary tissue present, the gland resembling in miniature the well differentiated adrenal of an adult.

It was desirable to determine whether the adult type of adrenal seen in such specimens has always been small or whether it has grown for a time and subsequently assumed a more mature type. The microscopic sections did not help in answering this question. However, the weights of the glands shown in the table, as well as the diagrams in figure 4, suggest that the pathologic process is probably prenatal atrophy. From the table it can be seen that with the exception of case 8, the adrenals weighed 1 Gm. or more in monsters weighing 2,000 Gm. or less. The weight of both adrenals never exceeded 0.8 Gm. in monsters more than 2,000 Gm. in weight. In other words, the smaller and presumably younger anencephalic fetuses had the larger amounts of

adrenal tissue, both by weight and size. This observation suggests that the adrenals had increased in size for a time before they assumed the small size usually found in the mature monsters.

RELATIONSHIP OF THE HYPOPHYSIS TO THE ADRENAL

Several conjectures have been offered to explain the changes in the adrenal with anencephaly. Alexander [14] regarded the adrenal lesion as

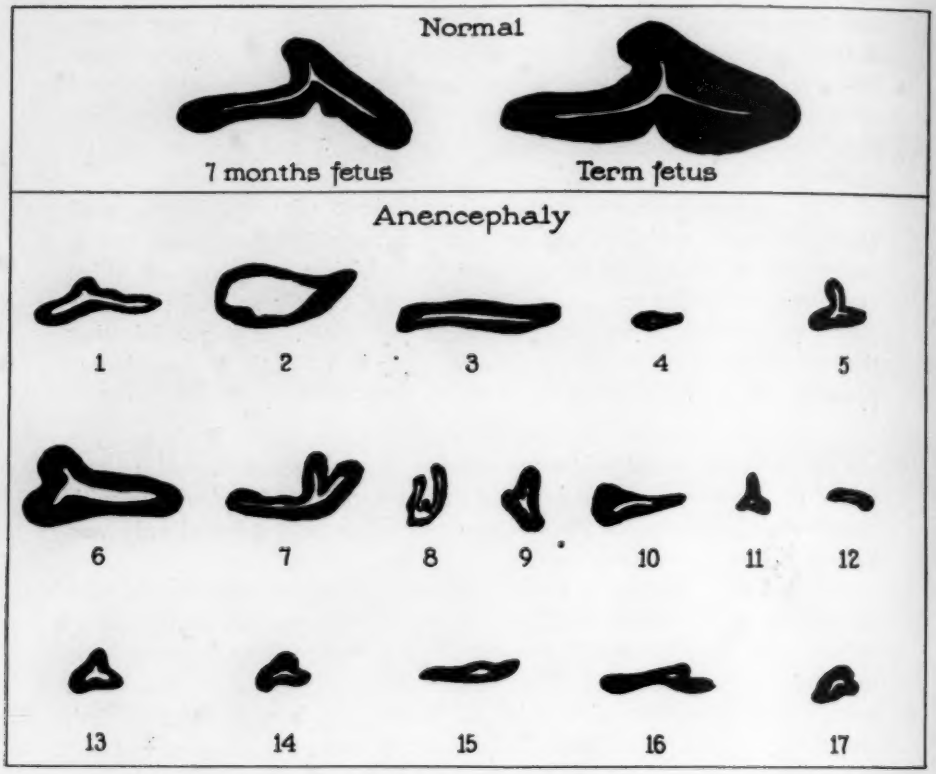


Fig. 4.—Drawings of sections of the adrenals from the same normal and anencephalic fetuses as shown in figure 2. The cortex is in solid black; the white area represents medulla. The glands are enlarged to about twice the normal size.

primary and the probable cause of anencephaly. Anomalous development is apparently not due to a defect in the sympathetic nervous system, for most observers have found that the adrenal medulla and chromaffin system are normal in anencephaly. Meyer [15] found that the adrenals of anencephalic fetuses developed normally until the fifth month of intra-

14. Alexander, C.: Beitr. z. path. Anat. u. z. allg. Path. **11**:145, 1892.
15. Meyer, R.: Virchows Arch. f path. Anat. **210**:158, 1912.

uterine life and therefore concluded that the adrenal lesion was secondary rather than primary. This suggestion corresponds with the observation that the smaller adrenals were found in the larger monsters.

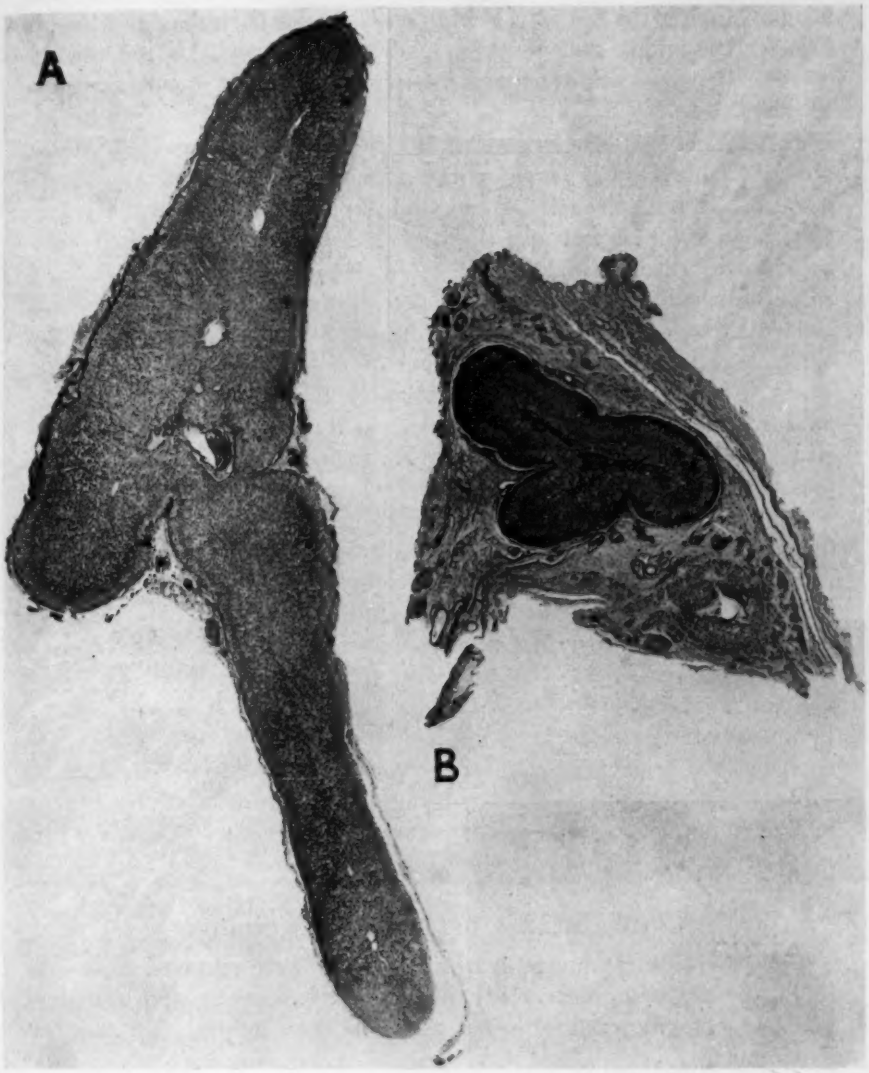


Fig. 5.—*A*, transverse section through the largest diameter of the adrenal of a normal fetus weighing 1,700 Gm.; × 10. *B*, transverse section through the adrenal of a monster (case 8) weighing 2,000 Gm. Note the differentiation of the cortical layer; × 10.

Kohn [6] and Moehlig [16] thought that the changes in the adrenal are secondary to those of the hypophysis. A careful examination of my cases does not support this conclusion, since some large hypophyses were associated with very small adrenal glands, which had excessive cortical atrophy, and vice versa. There was no demonstrable relationship between the middle and posterior lobes of the hypophysis and the size of the adrenal. Vaclav [10] found the same discrepancy in a larger series of cases.

The functional relationship of the two endocrine organs is defined with difficulty because anencephalic fetuses are continually under the influence of hypophysial hormones of the mother. Nevertheless, the experiments of several observers give support to the opinion that the change in the adrenal cortex is secondary to that in the hypophysis. Smith [17] demonstrated that hypophysectomy in rats caused rapid regression in the size of the adrenals. Houssay and Sammartino [18] observed similar cortical atrophy of the adrenals in hypophysectomized dogs. These experiments have been considerably clarified by the isolation of an adrenotropic factor by Collip, Anderson and Thomson [19] and of a corticotropic factor by Anselmino, Hoffmann and Herold.[20] Similar aplasia of the adrenal cortex is present both in dystrophia adiposogenitalis and in Simmonds' disease. It is believed to be secondary to a change in the anterior lobe of the hypophysis that interferes with the production of the adrenotropic hormone. It is noteworthy that with hyperplasia of the hypophysis, as in many cases of acromegaly, there is hyperplasia of the adrenal cortex. In view of these observations it is highly probable that the changes in the adrenal cortex with anencephaly are secondary to some alteration of the hypophysis or of its related structures induced by impaired development of tissues at the base of the brain. The anterior lobe being present in these cases, the hypophysial changes are probably due to some alteration in the normal function of the anterior lobe or in the normal relationship between the anterior lobe and the pars nervosa because of defect or absence of the latter.

OTHER GLANDS OF INTERNAL SECRETION

The thyroids, thymuses, ovaries and testes were removed from most of the fetuses with anencephaly, and histologic sections were compared with those from normal fetuses of about the same weight. No conspicu-

16. Moehlig, R. C.: Arch. Int. Med. **44**:339, 1929.

17. Smith, P. E.: J. A. M. A. **88**:158, 1927.

18. Houssay, B. A., and Sammartino, R.: Compt. rend. Soc. de biol. **111**:494, 1933.

19. Collip, J. B.; Anderson, E. M., and Thomson, D. L.: Lancet **2**:347, 1933.

20. Anselmino, K. J.; Hoffmann, F., and Herold, L.: Klin. Wchnschr. **12**: 1944, 1934; **13**:45, 1934.

ous changes were observed in the sections from the anencephalic monsters that were not present in those of normal fetuses.

CHOROID PLEXUS

Sections were made in several places from the hemorrhagic tissue overlying the base of the skull (fig. 1 *a*). It was very vascular, and in some specimens the epithelium was hyperplastic and often unassociated

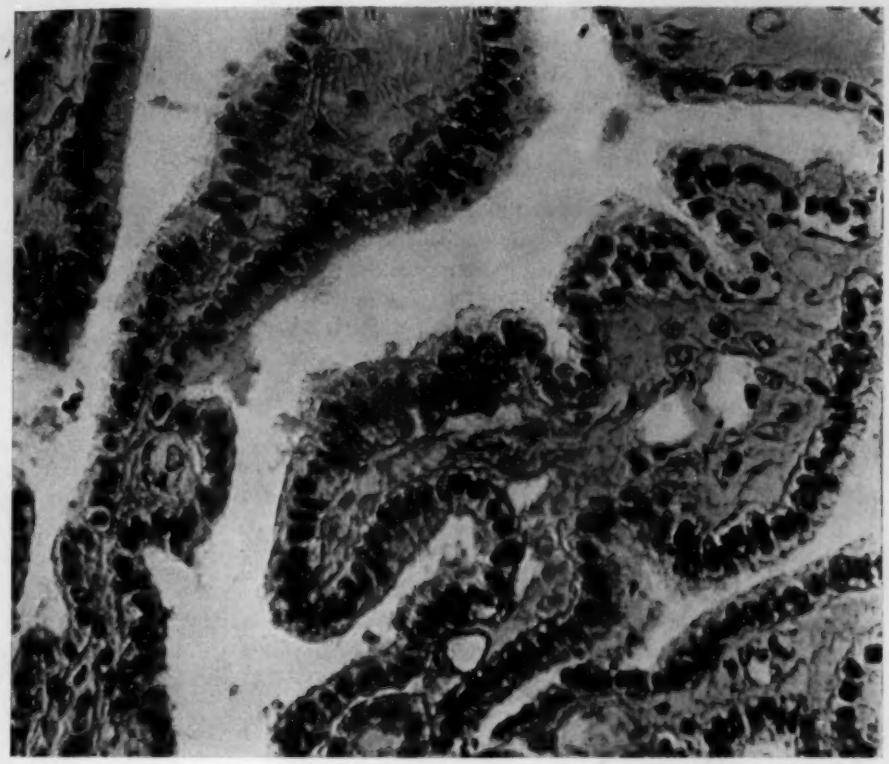


Fig. 6.—A section of hyperplastic vascular choroid plexus from the floor of the cranium of an anencephalic fetus weighing 2,000 Gm.; × 350.

with nerve tissue (fig. 6). Broder [12] described in a case of hydramnion a similar cuboidal epithelium over vascular capillary projections. Because of the frequent occurrence of hydramnion in anencephaly, an attempt was made to correlate this excess of fluid with the presence of exposed choroid plexus. In other words, is the hydramnion due to excessive secretion of cerebrospinal fluid? The lack of a well formed pia-arachnoid is probably important, for with a defect in this tissue even a normal amount of spinal fluid would not be reabsorbed. Choroid plexus tissue

was found in 9 of 13 cases examined (table). Of these 9 cases, a definite diagnosis of hydramnion, confirmed by roentgen examination, is recorded in 5 instances. In 1 case mention was made of the large size of the abdomen with no lightening, and in another a note had been made of "an abundance" of fluid. Of the remaining cases, there was no history available in 1 and no record of hydramnion in the other. In cases 3, 12 and 13 (table) there was no history of hydramnion, nor was any choroid plexus found. Only in case 1 was a diagnosis of hydramnion made although no choroid plexus tissue was found. The only cases in which there was choroid plexus with no hydramnion were cases 1 and 2, and since in these the fetuses were the smallest in the series, it seems possible that a slight excess of fluid may have been overlooked.

CONGENITAL ANOMALIES

Nine of 17 anencephalic monsters on which complete autopsies were performed had additional congenital defects. In 3 cases there was an incomplete fissure of the right lung; in 2, bilateral hydronephrosis, and in 1, unilateral hydronephrosis. Ovarian cysts were observed in 2 cases, an interventricular septal defect in 1 and Meckel's diverticulum in 1. One monster showed 4 additional anomalies, namely, a spina bifida, club-feet, a horseshoe kidney and an imperforate mouth.

SUMMARY

The anterior lobe of the hypophysis was found in all of 20 anencephalic monsters examined; the pars intermedia was found in 6 and the pars nervosa in 5.

The adrenals were found in all of 18 anencephalic monsters examined. The glands were all of the adult type because prenatal atrophy had apparently occurred in the developing adrenal tissue. The atrophy of the adrenals in anencephaly is probably the result of an impairment of the development of the hypophysis induced by changes in the tissues at the site of the base of the brain.

In most of the monsters the brain site was covered with hemorrhagic tissue in which there was a large amount of choroid plexus. It is suggested that this may be of some significance in the production of hydramnion.

Case Reports

OCCLUSION OF THE ENTIRE INFERIOR VENA CAVA

Report of a Case with a Review of the Subject

A. L. Sparks, M.D., and Herbert Fox, M.D., Cleveland

This paper presents a case of long-standing complete occlusion of the entire subdiaphragmatic portion of the inferior vena cava, with a review and discussion of the subject. In the case presented here, during a period of undetermined duration, an extensive collateral venous circulation had been established. While the patient was under clinical observation, progressive cirrhosis of the liver resulted in obstruction of the portal vein. The coexistence of complete occlusion of the inferior vena cava and partial obstruction of the portal vein constituted a baffling clinical problem and an extraordinary opportunity for clinical-pathologic correlation.

REPORT OF CASE

A 33 year old colored woman was admitted to the medical service three times. She first entered the hospital eighteen months prior to her death. She complained of shortness of breath and progressive edema of the lower extremities of a few weeks' duration. The present illness began a few weeks prior to admission, when she noticed shortness of breath on exertion and slight edema of the ankles. Her past history was noncontributory. During her six years of marriage she had only one pregnancy, and this terminated in a spontaneous abortion at three months, without known complications.

There was pitting edema of the entire lower extremities, extending upward to the level of the sacrum. There were signs of a small amount of fluid in the right pleural cavity and a moderate degree of ascites. The uterus and adnexa were firm and immobile.

The temperature was 38 C. (100.4 F.); the pulse rate, 100; the respiratory rate, 24, and the blood pressure, 110 systolic and 70 diastolic. The hemoglobin was 75 per cent; the red blood cell count, 4,000,000; the white cell count, from 3,000 to 5,700; the blood smear and differential count were normal. The urine and the renal function were normal. The electrocardiogram showed low voltage and slurring of the QRS complex in all leads. The total blood proteins amounted to 5.3 Gm. per hundred cubic centimeters; the albumin, to 2.76 Gm., and the globulin, to 2.55 Gm., per hundred cubic centimeters. The basal metabolic rate was normal.

After two weeks of rest in bed, receiving diuretics and digitalis, the patient lost 15 Kg. of weight, and only a slight degree of edema of the ankles remained.

An interval of one year elapsed before the second admission, which occurred six months prior to death. The patient enjoyed good health and was ambulatory, but on her visits to the outpatient department she complained of shortness of breath on exertion. Diuretics were used successfully for relief of her edema until a short while prior to the second admission.

From the Institute of Pathology, Western Reserve University, and the Department of Medicine, University Hospitals.

She was hospitalized the second time, because of enlargement of the abdomen and increasing edema of the lower extremities. She showed a marked degree of pitting edema below the waistline, bilateral hydrothorax, ascites of moderate degree, pelvic findings as noted previously and dilatation of superficial veins of the lower part of the abdominal wall. Biopsy of a large nodular vein near the umbilicus disclosed many dilated veins, some of which were filled with thrombi. The pathologic report was phlebectasia with thrombosis.

The hemoglobin was 78 per cent; the red blood cell count, 4,100,000; the white cell count, from 1,400 to 5,000. The Wassermann reaction of the blood was repeatedly negative. The blood protein averaged 4 Gm. per hundred cubic centimeters; the albumin, 1.68 Gm.; the globulin, 2.32 Gm.; the albumin-globulin ratio was 0.73.

The pleural and peritoneal fluids were repeatedly chylous.

The venous pressure in the arm veins was normal. In the femoral veins, the pressure was strikingly high. Readings of 35 cm. were repeatedly found. Fluoroscopic-roentgenographic examinations of the thoracic organs revealed that the mediastinum was fixed, that the cardiac pulsations were markedly diminished and that nodes in the tracheobronchial region were calcified.

During the four months of her hospitalization, the patient received a high vitamin, high protein diet, iron and liver therapy and roentgen radiation over the mediastinal and retroperitoneal lymph nodes. The pitting edema of the lower extremities and the moderate ascites persisted despite all therapeutic measures. There was a daily small elevation of temperature as well as a moderately rapid pulse while she was at rest in bed.

During an interval of two months at home, observation in the outpatient department showed no improvement. Ascites and increasing edema necessitated another admission to the hospital. This was twelve days prior to death. The results of physical examination, laboratory studies and studies of the venous pressure were similar to those previously recorded. The striking feature during the last three days of life was the rapidly progressive myocardial failure.

Various clinical diagnoses were considered. At the time of the first admission the condition was believed to be myocardial failure, origin unknown. Later, obstruction of the thoracic duct, nutritional edema, chronic adhesive pericarditis and chronic salpingitis were thought most probable. The diagnosis of obstruction in the inferior vena cava was made on the last admission, but until the autopsy was made, some form of heart disease was thought to exist. The obstruction of the inferior vena cava was thought to be due to tuberculosis of the mesenteric lymph nodes. The edema, the small quiet heart and the low voltage in the electrocardiogram were thought to be evidence of pericardial adhesions and cardiac compression. However, the venous pressure in the arm veins was not elevated, and the edema was limited to the lower extremities. The low level of the plasma proteins and the edema suggested nephrosis, but tests showed that the renal function was normal, and at no time was there albuminuria.

The evidence that led to a diagnosis of obstruction in the inferior vena cava was the persistent elevation of the venous pressure in the femoral veins, the dilatation of the superficial abdominal veins and the edema limited to the lower extremities and lower part of the abdominal wall. The majority of the clinicians who saw the patient did not think that the signs and symptoms could be produced by obstruction in the inferior vena cava or in the thoracic duct alone.

The autopsy (by A. L. S.) was made thirty minutes after death. External examination disclosed marked edema of the lower extremities, buttocks and abdominal wall. There were external hemorrhoids of moderate size. On section, 300 cc. of pale yellow fluid with a specific gravity of 1.010 was found in the peritoneal cavity, and 75 cc. of a similar fluid was found in each pleural cavity.

The inferior vena cava from below the hepatic veins to the common iliac veins was completely occluded and transformed into a solid fibrous cord. Throughout its course it was firmly attached to the prevertebral fascia. The renal veins were obliterated for a distance of from 2 to 3 cm. in direct continuity with the vena cava. A cross section of the vena cava showed only fibrous connective tissue, and no venous channel could be found. There was an organized thrombus, measuring 0.5 cm. in diameter, in the right common iliac vein. The omental blood vessels were distended, and the omentum was attached to the abdominal wall by fibrous adhesions.

The collateral circulation consisted of communications between the inferior caval tributaries and the portal, azygous, internal mammary and inferior phrenic veins. The left adrenal vein was greatly dilated, measuring about 2 mm. in diameter, and communicated with the left renal vein, splenic vein and left inferior phrenic vein. The right adrenal vein communicated with the right inferior phrenic vein. There were many communications between the ovarian veins and the mesenteric veins. Many aberrant renal veins were found which communicated with the mesenteric veins. There was an anastomosis between the hemorrhoidal plexus and the branches of the inferior mesenteric vein. Many communications were established between the branches of the internal mammary vein and the branches of the external iliac veins. The omentum which was adherent to the abdominal wall possessed many dilated veins, which were apparently in direct communication by means of vascularized adhesions with similar dilated veins in the anterior abdominal wall. The greatly dilated ascending lumbar veins emptying into the azygous vein were another important channel of venous return.

The liver weighed 1,020 Gm. and was coarsely nodular. There were three scars, measuring 2 cm. in diameter, on the anterior surface. The usual architecture was absent, and there was a marked increase in fibrous connective tissue in the portal spaces. The lungs showed small areas of bronchopneumonia and many small infarcts. There was a healed salpingo-oophoritis. The heart, spleen, pancreas and kidneys exhibited no gross pathologic change.

Microscopic examination of the vena cava showed it to be composed of dense, relatively acellular fibrous connective tissue. There was no vascular channel in the center, but there were a few small vascular spaces in the periphery which probably represented recanalization. Elastica stains showed an internal elastic membrane; in some sections it was intact and complete and extended around the inner portion of the fibrous cord, while in other sections it was fragmented and had no definite shape. The berlin blue reaction indicated no iron pigment.

Sections of the thoracic aorta showed syphilitic aortitis.

There was a moderate increase in fibrous connective tissue in the portal spaces in the liver, with many collections of lymphocytes and with proliferation of bile ducts and distortion of the architecture of the organ.

CLINICAL-PATHOLOGIC CORRELATION

The essential clinical features of this patient, who had been in good health eighteen months prior to death, were: persistent edema of the lower extremities, extending up to the lower level of the trunk;

chylous ascites and hydrothorax; markedly increased venous pressure in the lower extremities, in contrast with a normal pressure in the upper extremities; dilatation of veins of the lower abdominal wall; very low voltage and slurring of the QRS complex in the electrocardiogram.

The chief pathologic features were: organized occlusive thrombophlebitis of the inferior vena cava, with extensive collateral communication between the inferior caval tributaries and the portal, azygous and internal mammary veins; cirrhosis of the liver; bilateral healed salpingo-oophoritis.

The patient noticed no pronounced symptoms from the obstruction of the inferior vena cava until Laënnec's cirrhosis of the liver developed or advanced to such a degree that there was obstruction of the portal system. The communication between the inferior caval tributaries and the portal vein comprised the most important part of the collateral circulation, and when this was embarrassed by the cirrhosis of the liver, marked symptoms ensued. Owing to this process, the cirrhosis of the liver was the more serious disease and hastened the death of the patient.

The persistent edema and ascites could not be accounted for on the basis of cardiac or renal impairment or on that of endocrine disturbance. The dilatation of veins of the abdominal wall, studies of the results of venous pressure and the absence of edema in the upper half of the body, where the venous return was by tributaries of the superior vena cava, were evidence of venous obstruction located in the inferior vena cava system. Studies of venous pressure are to be considered valuable in the diagnosis of obstruction of portions of the caval system. In the cases of obstruction of the vena cava which have been reported in the literature, no mention is made of studies of venous pressure.

The ascites was probably produced by a combination of marked venous stasis in an overburdened portal system, cirrhosis of the liver and, in the later course of the disease, hypoproteinemia with an alteration in the albumin-globulin ratio. The diffusion of fluid containing proteins and metabolites into the tissue spaces and serous cavities contributed much to the persistence of edema, through loss of protein (of the albumin fraction particularly, because of smaller molecules) from the blood. The lowered albumin-globulin ratio and the decrease in total proteins contributed to cause the edema. This was shown by the development of transient edema of the upper half of the body when the patient was placed in a recumbent position for a period of twelve hours.

Chylous fluid by no means implies the presence of obstruction of the thoracic duct or rupture of a chyliferous vessel as presupposed by many observers. Blankenhorn,[1] in a study of 7 cases, found chylous fluid associated with a variety of conditions, including mediastinal tumor, nephritis and typhoid fever.

The fixed mediastinum was explained on the basis of the anchorage of the inferior mediastinal structures by the rigid inferior vena cava. The myocardium showed no abnormal changes. The changes in the electrocardiogram and the smallness and quietness of the heart were thought to be due to the delay in the venous return to the heart.

1. Blankenhorn, M. A.: Arch. Int. Med. **32**:129, 1923.

The syphilitic aortitis discovered at autopsy was not far advanced and produced no signs or symptoms. When the patient was first seen, there was no evidence of active inflammation in the pelvis. The pelvic organs were a possible source of the thrombophlebitis, but the inflammatory disease of the pelvis must have existed several months before the admission of the patient to the hospital.

ETIOLOGY

Pleasants [2] in 1911 collected 296 cases of complete or partial occlusion of the vena cava and added 18 cases from the Johns Hopkins Hospital. Two hundred and fourteen, or 72 per cent, were due to thrombosis. Other causes were pressure, diseases of the liver, neoplastic growth and embolism. In 90 per cent of the cases the thrombosis had its origin in the lower third of the vena cava, into which it had been propagated from the iliac veins. In most of the cases of occlusion by pressure there was also thrombus formation.

Since 1911, no similar complete report, with analysis of cases, has been made. Wakefield and Mayo [3] in 1934 reported a number of cases to demonstrate the effects of surgical ligation. The clinical features and the observations at autopsy were not discussed. A few reports of isolated cases have been found in the literature and also a number of cases have been listed in papers dealing with the general subject of thrombosis.

The antecedent or causal conditions in the cases collected by Pleasants [2] may be listed as follows:

	Cases
Puerperal sepsis	45
Neoplasms	83
Specific infectious diseases, including typhoid fever, tuberculosis and scarlet fever	42
Localized infections	17
Disease of the liver	31
Trauma	18
Congenital obstruction	9
Unknown causes	40
Other causes	29

In the majority of cases of obstruction reported in the literature, thrombosis was a very recent process and involved only a portion of the inferior vena cava, the distal portion being involved more frequently. In many there was only partial occlusion, and in only 2 cases was there fibrous obliteration as in our case. In these cases, the obliteration was below the renal veins, and it was thought to be congenital.

In the case reported here, the pathogenesis of the process is uncertain because the extensive fibrosis obscured all evidence of the primary disease. It was probably an old process, since there were no traces of iron pigment in the inferior vena cava. The spontaneous abortion which occurred six years previous to death may have been followed by a low

2. Pleasants, J. H.: Johns Hopkins Hosp. Rep. **16**:36, 1911.
3. Wakefield, E. G., and Mayo, C. W.: J. Missouri M. A. **31**:92, 1934.

grade sepsis, but there is no accurate information on this. The inflammation in the pelvis, which was represented at the time of autopsy by healed salpingo-oophoritis, may have been an etiologic factor. Thrombosis of infectious origin occurs most commonly following puerperal sepsis. However, in cases of the latter type septicemia and metastatic abscesses rapidly give a fatal termination. In this case syphilis cannot be incriminated by any evidence available.

Of 31 cases of obstruction of the vena cava in nonmalignant conditions of the liver, 10 were due to cirrhosis and 7 were due to syphilis. In none of these cases was there complete occlusion, and there was no edema or collateral circulation. Since cirrhosis was present in this case, there is a possibility that it may have been an etiologic factor. The cause of thrombosis associated with cirrhosis is obscure, although slowing of the venous blood flow plays a part. Since such an elaborate collateral circulation was established and no iron pigment was found in the remains of the vena cava, it is more probable that the occlusion of the vena cava preceded the development of cirrhosis of the liver.

SUMMARY

This report of a case of long-standing extensive obliteration of the inferior vena cava is combined with a discussion of other reports in the literature. Laënnec's cirrhosis of the liver was also found but was probably not the primary disease, and the cause of the venous lesion is obscure. Although certain of the clinical features suggested cardiac compression by pericardial disease, the study of venous pressures gave the principal clue to the correct diagnosis.

SYPHILIS OF THE HEART AND PERICARDIUM

EMMERICH VON HAAM, M.D., AND MICHAEL A. OGDEN, M.D.*
COLUMBUS, OHIO

The manifestations of acquired or congenital syphilis in the heart have been the subject of considerable discussion during the past twenty years. The majority of authors of the present day (Martland;[1] Saphir;[2] Clawson and Bell[3]) have been inclined to believe that infection with Spirochaeta pallida rarely produces anatomic lesions in the heart. While the significance of coronary changes, often observed in syphilitic persons, and of diffuse fibrosis of the myocardium are still disputed, the gummatous lesions of the heart are recognized as manifestations of syphilis, even though the spirochetal stains do not demonstrate any organisms in the majority of the cases. These lesions, which are rare, are usually described as diffuse or localized gummatous myocarditis (Sohval[4]). During a period of ten years Symmers[5] observed 4 cases of gummas of the heart in 4,880 autopsies at the Bellevue Hospital. Sohval found in the literature from 1845 to 1935 reports of 7 cases of diffuse gummatous myocarditis and 97 cases of "authentic" localized gummas of the heart. Among 5,213 autopsies in the years 1932 to 1935 at the State Charity Hospital of Louisiana, at New Orleans, we observed 3 cases of gummatous lesions of the heart which we shall report briefly.

REPORT OF CASES

CASE 1.—A 42 year old colored woman was admitted in September, 1932, complaining of a dull pain in the epigastric region, loss of appetite and vomiting after meals. Since the onset of her symptoms, four months before, the patient had lost about 70 pounds (31.8 Kg.) and had become so weak that she had to remain in bed. The history revealed nothing of importance; she had never been pregnant, and menstruation had ceased one and a half years previously. She was extremely emaciated. The heart and lungs revealed nothing that was abnormal. In the upper right quadrant of the abdomen was a tumor mass about the size of a lemon. Roentgen examination of the stomach showed a large filling defect in the pyloric region with marked gastric stasis. The gastric juice showed no free hydrochloric acid; there was occult blood in the feces. The Wassermann reaction of the blood was negative; chemical examination of the blood gave normal results. The diagnosis of gastric neoplasm was made. A severe psychosis developed, and the patient died shortly afterward. At autopsy (Dr. von Haam) the following diagnosis

*From the departments of pathology and bacteriology, Louisiana State University Medical Center and the State Charity Hospital of Louisiana, at New Orleans.

1. Martland, H. S.: Am. Heart J. **6**:1, 1930.
2. Saphir, O.: Arch. Path. **13**:266 and 436, 1932.
3. Clawson, B. J., and Bell, E. T.: Arch. Path. **4**:922, 1927.
4. Sohval, A. R.: Arch. Path. **20**:429, 1935.
5. Symmers, D.: J. A. M. A. **66**:1457, 1916.

was made: carcinoma of the stomach with metastases to regional, mediastinal and cervical lymph glands, and gummatous pericarditis.

The heart weighed 285 Gm. The heart muscle was brown and friable. The parietal and valvular endocardium did not show pathologic changes. The pericardium showed numerous yellowish white nodules from 0.5 to 2 cm. in diameter, extending somewhat into the myocardium. Between these nodules was a thick layer of partly organized fibrinous exudate, which in portions appeared hemorrhagic. The caseous character of the nodules was easily recognizable on the cut surface, through the pericardium. There was also an apparent increase in capillaries in the deeper pericardial fat tissue. The aorta showed only slight atheromatous changes. From the gross inspection, and particularly since the mediastinal lymph glands were extensively involved by the neoplastic process, a diagnosis of tumor metastasis to the pericardium was made. The true diagnosis was discovered only after microscopic examination of the specimen, at which time it could be demonstrated that the whitish nodules consisted largely of areas of caseous necrosis walled by newly formed capillaries and loose connective tissue. A few giant cells of the Langhans type were also found in the peripheral regions of these caseous areas. The entire pericardium was markedly thickened and showed granulation tissue with numerous capillaries and foci of round cell infiltration. Large collections of plasma cells were conspicuous, and the perivascular character of the round cell infiltrate was well marked. With consideration of the necrosis and of the presence of newly formed capillaries as well as of the large collections of plasma cells, the diagnosis of multiple gumma of the pericardium was made.

Case 2.—In April 1936, a 32 year old colored woman was admitted, with shortness of breath and swelling of the ankles. The dyspnea was particularly severe after exertion and at night, at which time she was unable to lie flat on her back. It had become so severe during the preceding month that she had been unable to do any work. She had had the usual childhood diseases, and the remaining history was irrelevant. She was markedly dyspneic, with slight peripheral venous congestion and vivid jugular pulsation. The blood pressure was 170 systolic and 60 diastolic, the pulse rate between 98 and 120 a minute and the respirations 22 a minute. The heart showed a displacement of the apex of 15 cm. from the midsternal line, toward the left, with the apex beat in the seventh intercostal space. There was marked palpitation, with pulsation visible over the entire area. Auscultation showed a "to and fro" murmur over the aortal and mitral orifices. There was beginning edema of the lungs, the liver was enlarged and tender, and the extremities showed marked chronic edema. The Wassermann reaction of the blood was anticomplementary on two occasions. The urine contained numerous granular and hyaline casts. The total red blood cell count was 3,750,000, with a normal white blood cell picture. The electrocardiographic examination showed a right ventricular predominance with T waves like those following administration of digitalis, a long PR interval and wide and notched PW waves. There was definite evidence of myocardial disease without disturbance in the cardiac rhythm. A clinical diagnosis of syphilitic heart disease was made, and the patient was given digitalis. After a short period of improvement she died suddenly, two weeks after admission. At autopsy (Dr. Ogden), the following diagnosis was made: gumma of the right side of the heart, involving the auricle and ventricle; syphilitic mesoaortitis with insufficiency of the aortic valves; saccular aneurysm of the ascending aorta; congestive heart failure.

The heart weighed 600 Gm. and measured 12.5 by 9.2 by 5.5 cm. The left ventricle measured 1.3 cm., the right 0.7 cm., in thickness. The pericardium showed numerous petechiae. Above the tricuspid valve in the wall of the right auricle,

and immediately below the valve in the wall of the right ventricle, several yellowish white nodules could be found protruding beneath the endocardium. They were of a soft elastic consistency and ranged from 0.5 to 2 cm. in diameter. The myocardium was pale and soft; the endocardium showed some subendocardial fat in the papillary muscles of the left ventricle. The commissures of the aortic valves were widened, and the leaflets were thickened and markedly shortened. The aorta showed numerous white edematous patches and deep parallel wrinklings. Four centimeters above the aortic valves there were two saccular pouches, each 2 cm. in diameter and about 1.5 cm. in depth. The walls of the pouches were thick and congested and showed nodular areas of necrosis, apparently small gummas. The microscopic examination of the nodules in the right ventricle showed extensive caseous necrosis, with no tissue structure recognizable in the necrotic area. There was a marked cellular infiltrate surrounding the necrotic area, chiefly of lympho-

Fig. 1 (case 2).—Gross specimen, showing multiple gummas in the intraventricular septum, protruding into the right ventricle of the heart.

cytes with an occasional neutrophil, eosinophil, leukocyte and plasma cell. There was some proliferation of capillaries and of fibrous tissue, the latter being composed of very young fibroblasts. The inflammatory process seemed strictly localized and did not extend into the surrounding tissue. The remaining myocardium showed moderate hypertrophy of the muscle fibers with no further changes. Sections through the aorta showed typical diffuse syphilitic aortitis. There were numerous small gummas in the walls of the small aneurysms.

Case 3.—A colored man 58 years of age had been repeatedly admitted to the hospital because of dyspnea on the slightest exertion, edema of the ankles and feet and cardiac asthma. At the present admission he also had a cough with blood-streaked sputum and frequent attacks of palpitation of the heart. He had marked cardiac decompensation. Dyspnea and edema of the lower extremities were pronounced. The blood pressure was 130 systolic and 84 diastolic, the pulse

rate 78 a minute and the respiratory rate 32 a minute. The apex impulse of the heart was in the seventh intercostal space, at the anterior axillary line. The heart beat was regular, and there was a rough systolic murmur over the mitral valve with sharply accentuated second arteria pulmonalis sound. The urine showed numerous granular casts. The patient died on the second day of admission. The clinical diagnosis was arteriosclerotic heart disease. At autopsy (Dr. Lafferty) the following diagnosis was made: arteriosclerotic heart disease with marked hypertrophy and dilatation of the left ventricle; possible gumma of the heart; syphilitic mesoaortitis; chronic congestive heart failure.

The heart was markedly increased in size and weighed 468 Gm. The epicardial surface in the region of the apex was hemorrhagic and soft. There were a few fibrous adhesions on the pericardium in this region. On section the left ventricle was markedly dilated. In the apical region there was an area measuring 2.5 cm. in which the wall was markedly thinned and dilated. There was marked fibrosis in this area. Situated in the posterior wall of the left ventricle near the base of the heart was a round area of yellowish white necrosis which was elastic, firm and surrounded by a very hemorrhagic grayish red zone of soft tissue resembling granulation tissue. The right auricle was also dilated. The coronary arteries were tortuous and showed narrowing of their lumens. The endocardium of the left ventricle showed a grayish white thickening, which in places had the appearance of a subendocardial exudate. The valvular endocardium did not show any changes. The aorta showed numerous areas of atheromatosis with small intervening zones of wrinkling.

The microscopic examination of the heart showed that at the apex, where the thinned-out area of myocardial fibrosis was found, there was marked fibrosis with considerable collagenous substance and only a small amount of muscle fiber in the fibrosed tissues. The surrounding muscle fibers showed marked hypertrophy. The deposition of hemosiderin pigment indicated an old myocardial hemorrhage, probably following the infarction. The arterioles of the surrounding muscle showed thickened walls but no destructive intimal changes or thrombi. The yellow area near the base showed extensive caseous necrosis, due to confluence of several small areas of necrosis. In the necrotic area all structure was lost. These necroses were walled off from the normal heart muscle by a dense network of dilated capillaries, which were surrounded by young fibroblasts and foci of small round cells. In a few places accumulations of endothelial cells similar to small tubercles, with one or more Langhans giant cells and surrounded by a wall of lymphocytes, could be found. However, even around these small areas of necrosis the new formation of capillaries was very evident. The areas of endocardial thickening described proved to be subendocardial granulation tissue, composed of newly formed capillaries, fibrocytes and small round cells. In some portions this granulation tissue extended into the papillary bundles of the myocardium but was never accompanied by necrosis. The aorta revealed mild but typical mesoaortitis with numerous perivascular nests of round cells. The diagnosis of arteriosclerotic heart disease and gummatous myocarditis was made. We considered the lesion at the apex to be a scar following an old myocardial infarction, while the lesion at the base of the heart was undoubtedly a localized gumma, similar to those observed in the second case. The granulation tissue in the subendocardial and adjoining myocardial layers was also thought to be of syphilitic origin, and this lesion was therefore classified as diffuse syphilitic endocarditis and myocarditis. With the "silver mirror method" devised by Steiner,[6] numerous

6. Steiner, G.: J. Lab. & Clin. Med. **23**:293, 1937.

spirochetes could be demonstrated in parallel bundles betwen the muscle fibers of the myocardium surrounding the gummatous lesion. Most of the organisms showed signs of degeneration, irregular spirals and nodular swellings; some of them, however, showed the unmistakable structure of S. pallida.

Study of these cases makes it evident that each represents a different type of syphilitic lesion of the heart. The patient in the first case, in whom no signs of heart failure were present before death, had

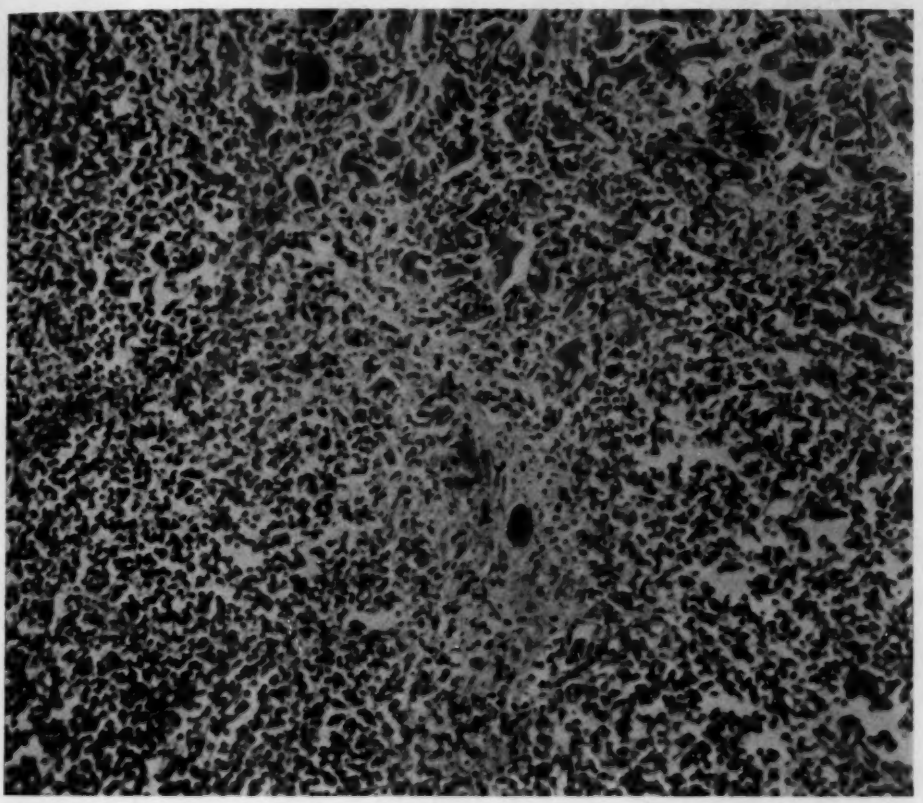


Fig. 2 (case 3).—Diffuse syphilitic myocarditis with numerous giant cells, plasma cells and fibroblasts.

gummatous pericarditis; the other two, who showed symptoms of failing heart before death, presented syphilitic lesions of the myocardium, the second revealing a localized gumma of the myocardium and the third a small gumma accompanied by diffuse syphilitic myocarditis and endocarditis. The remarkable feature of the first case is that the gummas were found in the pericardium, none being present in the myocardium, so that there was not the usual extension of the gummatous process from the myocardium into the pericardial tissue, which, according to Rosen-

thal,[7] is the most frequent cause of syphilitic pericarditis. Mönckeberg [8] believed that these "primary cases of gummatous pericarditis" were "great pathological rarities" and cited only 4 cases in which gummas of the pericardium were described with "some degree of reliability." We have not been able to find any such case recorded in the American literature.

The heart in the second case showed isolated gummas of the myocardium, typically located in the interauricular and interventricular septum. In spite of this location, no disturbance of the cardiac rhythm was noticeable, although the electrocardiogram showed definite evidence of myocardial damage. Attempts to find the spirochetes in the areas of gummatous necrosis were fruitless, a fact which, according to Schlesinger,[9] is typical for gumma following acquirement of syphilis, in contrast to gumma of the heart produced by congenital syphilis. The process was strictly localized, and the remainder of the heart showed only slight hypertrophy with little fibrosis but with marked degenerative changes. The heart failure was doubtlessly precipitated by the aortic regurgitation due to syphilitic changes in the aortic valves.

The third case is the most interesting of the three. Similar to that of Reifenstein,[10] it presented the acute clinical course of myocardial infarction, and at autopsy there was difficulty in differentiating the lesions from myocardial infarcts. Only the yellow color and the peculiar configuration of the area at the base of the heart caused us to suspect the correct diagnosis. The histologic examination showed, aside from the localized small gumma of the cardiac base, a rather ill defined granulomatous tissue, which, we later noted, occurred within the myocardium as a firm yellowish white wavy zone, quite similar to the change found in the case of Berblinger.[11] This tissue consisted of granulation tissue rich in giant cells, epithelioid cells and capillaries and extended diffusely through the myocardium. Numerous spirochetes could be demonstrated in the myocardium close to the granulomatous lesion. In the endocardium at the corresponding site, a similar granulation tissue was observed, which was also grossly recognizable as a whitish gray thickening similar to that described by Fischer.[12] Our observation suggests that such an inflammatory process in the endocardium is merely a continuation of the myocardial disease and does not represent a separate manifestation as suggested by Ribbert.[13] Diffuse gummatous myocarditis is extremely rare, and Sohval [4] reported that only 7 instances were recorded in the literature. However, it seems to occur more fre-

7. Rosenthal, O.: Berl. klin. Wchnschr. **37**:1081, 1900.

8. Mönckeberg, J. G., in Henke, F., and Lubarsch, O.: Handbuch der speziellen pathologischen Anatomie und Histologie, Berlin, Julius Springer, 1924, vol. 2, pp. 453 and 587.

9. Schlesinger, H.: Syphilis und innere Medizin, Vienna, Julius Springer, 1928, p. 3.

10. Reifenstein, E. C.: Ann. Int. Med. **10**:241, 1936.

11. Berblinger, W.: Centralbl. f. allg. Path. u. path. Anat. **21**:1045, 1910.

12. Fischer, B.: Frankfurt. Ztschr. f. Path. **7**:83, 1911.

13. Ribbert, H., in Henke, F., and Lubarsch, O.: Handbuch der speziellen pathologischen Anatomie und Histologie, Berlin, Julius Springer, 1924, vol. 2, p. 244.

quently in combination with localized gumma than in its pure form (Mönckeberg [8]). The thinned-out area in the apex of the heart was probably an old myocardial infarct, as the autopsy showed considerable changes in the wall of the coronary vessels, similar to the ones described by Benson [14] and by Cowan and Faulds.[15] Although Levine [16] expressed the belief that syphilis is a common cause of coronary thrombosis, Saphir [2] correctly suggested that mere coincidence of both conditions does not prove any direct causative influence. In our case extensive arteriosclerotic changes of the entire vascular apparatus were present and, as far as the aorta was concerned, overshadowed the syphilitic manifestations.

SUMMARY

Three cases of syphilitic gumma are reported. The first was a case of isolated gummatous pericarditis; the second, a case of multiple gummas in the myocardium, and the third, a case of a small gumma accompanied by extensive diffuse gummatous myocarditis and syphilitic endocarditis.

14. Benson, R. L.: U. S. Vet. Bur. M. Bull. **5**:13, 1929.
15. Cowan, J., and Faulds, J. S.: Brit. M. J. **2**:285, 1929.
16. Levine, S. A., and Brown, C. L.: Medicine **8**:245, 1929.

Laboratory Methods and Technical Notes

A TRAY FOR DEHYDRATION, CLEARING AND PARAFFIN IMPREGNATION OF LARGE QUANTITIES OF TISSUE

HAROLD WOOD, M.D., CHARLESTON, S. C.

This tray was designed to eliminate the time and effort required to transfer blocks of tissue through a series of bottles containing dehydrating and clearing agents and receptacles containing "soft" and "hard" paraffin. The idea of such a tray is not new, but the design of this one furnishes an inexpensive durable apparatus consisting of a single unit of large capacity which is not subject to the possible uncertainties of operation of intricate mechanical and electrical devices.

Such a tray can be constructed by any competent machinist [1] or sheet metal worker, and the design may be modified to suit the needs of the individual laboratory. The cost of materials for the one to be described was approximately $12.

DESCRIPTION OF TRAY

Specifications and Assembly.—Except for the legs and the handle, the tray was constructed entirely of perforated sheet Monel metal, of which approximately 220 square inches (1,419 sq. cm.) is required. A standard sheet 14 by 26 inches (35.5 by 66 cm.) of 26 gage, with 225 holes per square inch (6.5 sq. cm.) may be purchased through local dealers. The perforations are 1/16 inch (0.16 cm.) in diameter and are spaced at a similar distance. Approximately 15 inches (38 cm.) of 1/8 inch (0.32 cm.) stainless steel rod are required for the four legs and handle (figure).

The diameter of the tray is 7½ inches (19 cm.) and the depth 2 inches (5 cm.). To the inner surface of the upturned edge of the circular disk which forms the base, the outer wall of the tray is soldered. One and one-half inches (3.8 cm.) inside the outer wall is a concentric partition of similar height (2 inches), and 1¼ inches (3.2 cm.) inside this partition is a second. Two cross partitions are fitted together at right angles to each other and into the two concentric circular partitions by cross lap joints, and they are soldered together, to the circular partitions, the outer wall and the base. This produces a central part divided into quadrants and two concentric troughs, each divided into four equal parts. Each fourth of the outer trough is divided into five compartments of equal size by radially arranged vertical partitions soldered into position. Similarly, each fourth of the inner trough is divided into three compartments. This produces a total of thirty-six compartments.

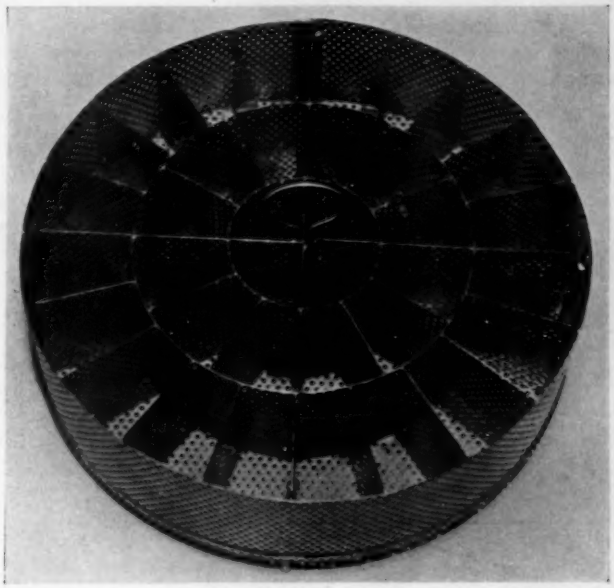
The legs elevate the base of the tray ½ inch (1.3 cm.). The rods forming the legs perforate the base, and one leg is soldered into one of the two angles

From the Department of Pathology, Medical College of the State of South Carolina.

1. This tray was constructed by Mr. C. D. Coleman, laboratory engineer of the Medical College of the State of South Carolina.

formed by each junction of the two main cross partitions with the outer wall. The handle is formed from a loop of the stainless steel rod, the ends of which are soldered into opposite angles formed centrally by the two main cross partitions.

Convenient Procedure and Equipment for Using the Tray.—The dehydrating and clearing solutions are contained in preservative jars with straight sides and ground glass covers. The latter prevent excessive evaporation of the fluids. The jars employed here are 8 inches (20.3 cm.) in diameter and 6½ inches (16.5 cm.) deep (inside measurements). Each requires about 1,500 cc. of fluid to immerse the tray to within a half inch of its top. Each dehydrating and clearing agent may be used for about one week without change. The percentage of the alcohols



Photograph of tray viewed obliquely from above.

may be judged with an alcohol hydrometer, but after some experience the state of the solutions can be judged by their appearance and by the consistence of the tissue when cut on the microtome.

Three pounds of each paraffin are required to immerse the tray sufficiently, and each batch may be used at least three weeks. The "soft" paraffin is contained in an uncovered glass preparation dish of slightly greater diameter than the tray and is kept in the paraffin oven at about 54 C. For the "hard" paraffin, in this laboratory, an enamel ware double boiler is used, the upper compartment of which is 9 inches (22.8 cm.) in diameter and 3½ inches (8.9 cm.) deep (inside measurements). The paraffin is placed in the uncovered upper compartment, and the lower compartment is kept partially filled with water. The assembled double boiler remains in the paraffin oven at about 58 C. except while the tissues are being embedded, when it is removed and placed on a previously heated electric plate. The water is thus kept hot enough to prevent the paraffin from cooling and hardening. The tray remains in the paraffin until all the tissue blocks have been embedded. It is then removed, drained and placed in a covered preservative jar

containing waste xylene, which removes the excess of paraffin. Before using the tray again, it has been found advantageous to wash it in waste 95 per cent alcohol.

The consumption of dehydrating and clearing solutions has been considerably reduced since this tray was put into use, and the consumption of paraffin is about as it was formerly.

For marking the tissue blocks ordinary marking tags are used, and the case numbers are written with a no. 2 lead pencil. The tags are carried through all of the solutions with their respective tissues, and the numbers remain legible.

Most of the material arrives at the laboratory already fixed in solution of formaldehyde (10 per cent concentration of U. S. P. solution); otherwise, the tray would probably be used for fixing the tissue. Small fragments of tissue, such as uterine curettings, may be wrapped in gauze, and with autopsy or large specimens from which numerous blocks of tissue are taken, the blocks may be distributed through two or more compartments in the tray to insure adequate exposure to the solutions.

In this laboratory specimens are examined from approximately 7,000 surgical operations and 225 autopsies yearly. This tray has been in constant use for six months, and it readily accommodates that volume of material. The use of the tray has not altered the high quality of the histologic sections.

General Review

SUBDURAL HEMATOMA

A. B. BAKER, M.D.
MINNEAPOLIS

In spite of the extensive literature that has accumulated on the subject of the subdural membrane, there still remain many uncertainties regarding its causes and pathogenesis. It was with the intent of investigating these phases of the subject that a review of the literature and a detailed study of the material available at the University of Minnesota was undertaken.

ETIOLOGY

The cause of subdural hematoma is at present a moot question. There has been within recent years a tendency to assign every hematoma to the category of trauma. However, one must acknowledge a spontaneous hemorrhagic and inflammatory as well as a traumatic form, since it is certain that in many cases trauma of even a mild degree plays no part.

In the middle of the nineteenth century there were two distinct schools of thought concerning the etiology of this condition. Virchow [1] and Heschl [2] believed that an inflammatory process within the dura caused primary formation of a membrane and produced the foundation for a hemorrhage, which was secondary. This view was rapidly accepted by such investigators as Schuberg,[3] Lancereaux,[4] Kremiansky [5] and Rindfleisch.[6] The English school questioned the primary inflammatory origin of the membrane. They believed that in most of the cases the subdural membrane was the result of a primary extravasation of blood into the subdural space. The cause of this bleeding was variously attributed to venous congestion, "dry cupping" from cerebral atrophy, degeneration of blood vessels, various systemic conditions with injury

From the Division of Nervous and Mental Diseases, University of Minnesota.

1. Virchow, R.: Verhandl. d. phys.-med. Gesellsch. **7**:134, 1857.

2. Heschl, R.: Compendium der allgemeinen und speziellen pathologischen Anatomie, Vienna, Wilhelm Braumüller, 1855, p. 289.

3. Schuberg, W.: Virchows Arch. f. path. Anat. **16**:464, 1859.

4. Lancereaux, E.: Arch. gén. de méd. **2**:526, 1862.

5. Kremiansky, J.: Virchows Arch. f. path. Anat. **42**:129, 1868.

6. Rindfleisch, E.: A Manual of Pathological History, London, New Sydenham Society, 1872, p. 302.

to vessels, diseases of the blood and trauma with rupture of vessels. This view was accepted and elaborated on by many investigators.[7]

Exudative Origin.—The chief advocate of the theory that the subdural hemorrhage results secondary to some inflammatory process of the dura was Virchow.[1] He believed that the dura becomes chronically inflamed and hence covered over its inner surface by a fine fibrinous exudate, sometimes blood tinged. Into this exudate the widely scattered dural vessels send capillaries, and their proliferation results in a vascular tissue composed of extremely thin-walled, irregular, wide, anastomosing vessels. This vascular membrane later becomes fibrous and adheres more and more to the inner surface of the dura. If the inflammation recurs, another layer of fibrin may form on the inner surface of the newly formed membrane and undergo organization. Virchow claimed to have seen cases in which there were from five to twenty layers in the pseudomembrane. Under conditions of hyperemia the newly formed vessels of the subdural membrane may rupture, producing small ecchymoses, which are absorbed, leaving many hemosiderin granules scattered between the dural fibers. In some cases many of the capillaries tear and produce massive hemorrhages, which in turn may organize and their capillaries rupture. To the fibrinous exudation and its organization Virchow gave the name "pachymeningitis interna chronica." The hemorrhagic membrane he called "pachymeningitis haemorrhagica." Since Virchow's description numerous confirmatory reports have appeared, all emphasizing the importance of a primary dural change as the cause of the subdural hemorrhage.

In 1860 Klob [8] described a hemorrhagic inflammatory lesion of the dura as a result of syphilitic infection. Wagner [9] in 1868 studied an 8 year old child with scurvy, who at autopsy revealed old inflammatory changes in the meninges, with the formation of a vascular cellular

7. (*a*) Baillarget, M.: Arch. gén. de méd. **5**:86, 1834. (*b*) Sperling, H.: Centralbl. f. d. med. Wissensch. **29**:449, 1871. (*c*) Huguenin, G., in von Ziemssen, H.: Cyclopedia of the Practice of Medicine, New York, William Wood & Company, 1877, vol. 12, p. 386. (*d*) Wiglesworth, J.: J. Ment. Sc. **33**:509, 1887-1888. (*e*) Robertson, G. M.: ibid. **39**:203, 1893. (*f*) Köhl, E.: Kor.-Bl. f. schweiz. Aerzte **23**:783, 1893. (*g*) Heubner, O.: Virchows Arch. f. path. Anat. **84**:267, 1881. (*h*) Buss, O.: Ztschr. f. klin. Med. **38**:451, 1899. (*i*) van Vleuten, C.: Ueber Pachymeningitis haemorrhagica interna traumatica, Bonn, C. Georgi, 1898. (*j*) Kasemeyer, E.: Deutsche med. Wchnschr. **43**:2020, 1912. (*k*) Trotter, W.: Brit. J. Surg. **2**:271, 1914. (*l*) Aubanel, M.: Ann. méd. psychol. **2**:55, 1843. (*m*) Boudet, C. E.: Mémoire sur l'hémorrhagie des méninges, Paris, Éverat & Cie, 1838. (*n*) Lélut, F.: Gaz. méd., Paris **4**:2, 1836. (*o*) Laborde, M.: Compt. rend. Soc. de biol. **24**:13, 1872.

8. Klob, J.: Wien. med. Wchnschr. **8**:116, 1860.

9. Wagner, B.: Jahrb. f. Kinderh. **1**:106, 1868.

membrane. He believed that such newly formed vessels frequently ruptured, producing a secondary hemorrhage. Alcoholism as a cause of dural changes with associated hemorrhage was advocated by Kremiansky [9] in 1868 and by Moses [10] in 1873.

Wohlwill [11] reviewed 30 cases of hemorrhagic pachymeningitis. He concluded that this was primarily a growth of the subendothelial tissue, probably resulting from a lesion of the endothelium (dural). He believed that numerous mechanical and chemical irritants could cause the dura to react with an endothelial and vascular proliferation. The fibrinous, serous and hemorrhagic processes were accidental features accompanying this endothelial injury and subendothelial proliferation. Jores and Laurent,[12] Fahr [13] and Rosenberg [14] also believed that the hemorrhagic membrane was the result of a primary growth of the capillaries and of the surrounding stroma of the inner part of the dura. Schwartz [15] emphasized the possible infectious nature of the dural membrane. He suggested that pachymeningitis as it occurs in infancy is an infectious disease and may be caused by the meningococcus or other organisms. He reported the occurrence of such a lesion in a 6 month old child, from which meningococci were isolated. Rosenberg,[14] on the other hand, believed that pachymeningitis haemorrhagica interna made the dura especially prone to meningitis. He reported 9 deaths from meningitis and insisted that the meningitis was secondary to the new membrane formation.

Numerous reports have been published of cases in which the authors have found a chronic inflammatory process of an undetermined nature which they believed preceded the dural hemorrhage. Mettenheimer [16] in 1868 reported such a finding in a 17 month old child. At autopsy a newly formed membrane covered the hemispheres, falciform ligament and tentorium. The hematoma contained 1½ quarts (1,400 cc.) of a dark liquid. Probst [17] reported the interesting case of a 15 year old patient in whom a chronic inflammation of the dura, involving both brain and cord, was found. In the cord there was only an inflammatory membrane, while in the brain there was definite hemorrhagic pachymeningitis. This report is one of the most convincing to prove the true

10. Moses, S.: Jahrb. f. Kinderh. **4**:152, 1873.

11. Wohlwill, F.: Virchows Arch. f. path. Anat. **214**:388, 1913.

12. Jores, L., and Laurent, H.: Beitr. z. path. Anat. u. z. allg. Path. **29**:486, 1901.

13. Fahr, T.: Centralbl. f. allg. Path. u. path. Anat. **23**:977, 1912.

14. Rosenberg, O.: Klin. Wchnschr. **49**:2272, 1913.

15. Schwartz, A. B.: Am. J. Dis. Child. **11**:23, 1916.

16. Mettenheimer, C.: J. f. Kinderkr. **50**:96, 1868.

17. Probst, M.: Arch. f. Psychiat. **36**:114, 1903.

inflammatory nature of at least some of the hemorrhagic subdural membranes.

Gordon [18] described 2 cases of pachymeningitis. In one there was a hemorrhagic, while in the other there was a nonhemorrhagic, membrane. He believed that numerous conditions, such as syphilis, tuberculosis, scurvy and the infectious diseases, may play a part in the production of the primary dural changes.

From this review it becomes readily apparent that in many cases so-called subdural hematoma is really a secondary hemorrhage into an inflammatory membrane which has appeared subsequent to an acute or chronic dural infection or irritation. In such cases the name originally applied to it by Virchow—"pachymeningitis haemorrhagica interna"—is most appropriate and descriptive.

Hemorrhagic Origin.—Much has been written on the belief that subdural hematoma is primarily a hemorrhagic process with a secondary dural reaction as a result of stimulation from the blood. It is suggested that the first development in the formation of the membrane is a primary bleeding into the subdural space. The effused blood coagulates, and a fibrinous membrane forms, enclosing the more fluid portion, which is gradually absorbed. The fibrinous portion, containing much blood pigment, becomes organized by capillaries and fibroblasts growing out from the dura. The subsequent changes in this membrane are much the same as in any organization. The chief problem is to decide just what is the cause of the original hemorrhage. As was stated before, the most recent trend is to acknowledge trauma as the primary source of the bleeding, even though the elicited history may in many cases point to an injury ridiculously trivial.

One of the chief arguments in favor of the traumatic causation is based on the normal anatomy of the cerebral veins. The superficial cerebral veins have a wide distribution. Some pass anteriorly in the direction of the frontal pole; others pass superiorly to the superomedial margin of the hemisphere, while the remainder pass posteroinferiorly to the temporo-occipital region. The majority of these veins terminate by passing from the surface of the cerebrum into one of the large venous sinuses, either the superior sagittal or the transverse. Most of the veins passing from the brain to the tributaries of the longitudinal sinus are short trunks and extend directly from the brain to the dura at right angles, with no tortuosity to allow for possible movement of the brain. Such veins have their cranial ends firmly fixed by the rigid dura, while the cerebral ends are attached to the movable hemisphere. The falx

18. Gordon, A.: New York State J. Med. **99**:720, 1914.

protects the brain from transverse displacement, but there is no protection against anteroposterior movement. Hence a sharp blow on the front or on the back of the head can easily produce a good deal of cerebral dislocation with rupture of these veins.

Recognition of trauma as a cause of subdural hematoma dates back to a report by Bouillon-Lagrange [19] in 1847 and to subsequent early reports by Schuberg,[3] Herter,[20] Göppert [21] and Finkelstein.[22] Bouillon-Lagrange reported the case of a 70 year old man in whom a subdural hematoma occurred following definite severe cerebral trauma. Herter mentioned the etiologic factor of trauma, but in the cases of his patients, 2 infants, absolutely no history of trauma was obtained. Göppert studied the cases of 3 children. He believed that malnourishment may influence the course of the disease but that trauma initiates the hematoma.

In more recent times the influence of trauma in this condition has been emphasized by Trotter,[7k] Kowitz,[23] Jelsma,[24] Frazier [25] and others. Trotter believed that most subdural hematomas had been traumatic in origin, some resulting even from slight traumas. He reported 4 cases in only 1 of which the hematoma was definitely of a traumatic nature. In the other cases the history of injury was indeed slight and of questionable significance. He suggested that once the hemorrhage has occurred, the enclosing membrane is derived from the organization of the outer layer of clots. After this membrane has formed, the tendency of the blood to coagulate within is checked. The enclosed collection of blood, which usually surrounds the torn veins from which it comes, remains fluid and delays thrombosis of these injured vessels. In this way the encysted hemorrhage continues to communicate with the vein, while its capsule affords enough resistance to prevent too rapid an increase in size.

Gardner [26] agreed with the aforementioned author as to the cause of the primary hemorrhage, that the bleeding is a result of a tearing of veins as they enter the sinus. However, he disagreed as to the cause of the secondary increase in size of the hematoma. He explained the increase in size after a latent period as follows: The original hematoma becomes clotted, and within a few days the clot is surrounded by a

19. Bouillon-Lagrange: Arch. gén. de méd. **14**:313, 1847.
20. Herter, C. A.: Am. J. M. Sc. **116**:202, 1898.
21. Göppert, F.: Jahrb. f. Kinderh. **61**:51, 1905.
22. Finkelstein, H.: Jahrb. f. Kinderh. **74**:451, 1911.
23. Kowitz, H. L.: Virchows Arch. f. path. Anat. **215**:233, 1914.
24. Jelsma, F.: Arch. Surg. **21**:128, 1930.
25. Frazier, C. H.: Ann. Surg. **101**:671, 1935.
26. Gardner, W. J.: Arch. Neurol. & Psychiat. **27**:847, 1932.

capsule of connective tissue growing out of the dura. The encapsulated clot undergoes partial liquefaction, the resultant fluid being high in protein content and separated from the subdural space merely by the thickness of a few layers of cells constituting the cyst wall. Since this membrane is impermeable to large protein molecules in the hemorrhagic fluid, an osmotic imbalance in favor of the hemorrhagic fluid must exist, resulting in the drawing of cerebrospinal fluid into the cyst. Therefore the cyst content increases in quantity even though the original hemorrhage has ceased.

Kowitz [23] studied 5,998 children under 2 years of age in whom intracranial hemorrhages had occurred. In 55.2 per cent the bleeding was in the dura. The author believed that in infants pachymeningitis haemorrhagica interna results from long hard births and rapid births, in both of which trauma plays an important role. Jelsma [24] recorded 2 cases in which injury produced dural bleeding. He believed that in many cases the injury is of such slight degree as to cause little immediate damage. Frazier [25] studied 6 cases, in 3 of which trauma was accepted as the determining factor. In 3 cases there was no history of injury of any type.

Putnam and Cushing [27] reemphasized Trotter's idea of the causation of traumatic subdural hemorrhages. They believed that the source of the hemorrhage could be found in a rupture of a sinus, of its lacunae or of tributaries of veins of the pia to the sinuses, and that apparently insignificant trauma to the head may in certain persons be followed by such an injury and subdural hemorrhage. Keegan,[28] Naffziger and Brown,[29] Leary [30] and Pedersen [31] all advocated a traumatic origin of the subdural hemorrhage. Keegan in 1 of his cases observed an opening in the superior longitudinal sinus which was the source of the hematoma. Leary stated that relatively trivial blows or falls on the head which have been experienced previously by a victim of subdural hematoma without harmful effects may be responsible for hemorrhage at the critical moment. He recorded severe injuries in a little over half his cases. In 8 cases he found a bleeding vein, and in 5 of these it was a vein bridging the dura.

There have been a few investigators who have cautioned against too widespread an acceptance of the traumatic origin of subdural mem-

27. Putnam, T. J., and Cushing, H.: Arch. Surg. **11**:329, 1925.

28. Keegan, J. J.: Arch. Surg. **27**:629, 1933.

29. Naffziger, H. C., and Brown, H. A.: S. Clin. North America **14**:1465, 1934.

30. Leary, T.: J. A. M. A. **103**:897, 1934.

31. Pedersen, O.: Deutsche Ztschr. f. Nervenh. **138**:229, 1935.

branes. Boekmann [32] studied 57 cases of operation on the brain, with blood appearing on the inner side of the dura as a result of the operation. In none were there microscopic signs of dural inflammation. There was no formation of a subdural membrane. He believed that bleeding alone results in absorption of the blood but not in formation of a membrane. He insisted that in healthy people trauma and bleeding alone are insufficient to produce pachymeningitis haemorrhagica interna. He was certain that trauma had been overemphasized. Sherwood [33] reviewed 9 cases of chronic subdural hematoma in children. No history of trauma was elicited in any of these cases.

It is true that in many cases trauma may play an important role as the primary cause of a subdural hemorrhagic membrane. But one must be guarded in insisting that in all cases such a membrane is due to injury and then accepting many too trivial injuries as an important factor in this condition. It is plain from a review of the literature that other causal factors may be found to account for many of the membranes. These factors result from injuries of the dural blood vessels with associated bleeding, as may be expected in the hemorrhagic diathesis or in acute and chronic infectious diseases with injury of vessels and bleeding, as well as in injury of vessels secondary to alcoholism or to terminal degeneration as seen in atherosclerosis.

Robertson [7e] believed that the cause of subdural hemorrhage is a sudden lowering of intracranial pressure, producing an effect analogous to "dry cupping" of the dura. He contended that this condition is most frequent in the patient with dementia paralytica and the senile person, in whom the brain is atrophied. Such persons are subject to great cerebrovascular disturbances and to sudden congestive or apoplectiform attacks which must disturb intracranial pressure. A sudden shrinking of the brain from a sudden vasomotor spasm, with anemia, or from a great fall of arterial pressure in an exhausting disease, whereby a deficient amount of blood is sent to the brain, prevents the compensatory mechanism from functioning and produces a condition allied to "dry cupping." This lessened intracranial tension disturbs the normal balance between intravascular blood tension and the tone of the vessels. The blood tension overcomes the strength of the walls of vessels, and a great dilatation of the vessels and engorgement with blood ensue. The inner surface of the dura becomes covered with engorged, dilated and bleeding vessels. Vessels will be seen with aneurysmal dilatations, as if distended to the bursting point. Minute hemorrhages can be found in many areas.

32. Boekmann: Virchows Arch. f. path. Anat. **214**:380, 1913.

33. Sherwood, D.: Am. J. Dis. Child. **39**:980, 1930.

If the intracranial tension is not balanced, the overdistended vessels rupture to produce massive hemorrhages. The blood coagulates and then organization occurs in a normal manner. In an old membrane, therefore, hemorrhages of all ages may be found, some recent and resembling a blood clot, some discolored.

Rosenberg [14] suggested that the hemorrhage in pachymeningitis haemorrhagica interna is due to thrombosis and venous congestion resulting in hyperemia and rupture of the dural vessels. He held that the cavernous sinus becomes thrombosed as a result of an infection or of a marantic condition. The veins emptying into the sinus, namely, the middle meningeal, the ophthalmic, the central retinal, the ethmoid and the supraorbital, become obstructed and congested. Congestion of the ophthalmic vein causes retinal hemorrhages, while involvement of the middle meningeal vein results in dural congestion and hemorrhage. He was not able, however, in spite of a careful search, to find such a thrombosis in any of his cases.

Russell and Cairns [34] reported 4 cases of metastatic neoplastic infiltration into the veins of the dense dural layer resulting in engorgement and rupture of the vessels in the areolar layer, with diffuse extravasation of blood into both the areolar dura and the subdural space. They explained this subdural bleeding on the basis of the following anatomic facts: The areolar layer of the dura immediately adjacent to the endothelial lining of the subdural space contains a rich network of capillaries. These drain outward into a second plexus in the dense dura and thence either to the periosteum of the calvaria or to veins that accompany the meningeal arteries. Therefore, obstruction of veins in the outer dense layer of dura will produce dilatation of capillaries in the inner areolar layer with secondary rupture and hemorrhage.

Sutherland [35] reported 2 cases of hemorrhagic pachymeningitis in infants, associated with scurvy. Frazier [25] observed a patient with a subdural hematoma who had frequent attacks of epistaxis. He wondered if the condition could be interpreted as evidence of a blood dyscrasia.

Reimer [36] and Hada [37] recorded cases of subdural hematoma associated with whooping cough. They believed that the toxin of the disease weakened the dural vessels and that then the whooping cough caused them to rupture by momentarily stopping the outflow of blood from the cerebral vessels, thus increasing the pressure in these vessels.

---

34. Russell, D. S., and Cairns, H.: Brain **57**:32, 1934.
35. Sutherland, G. A.: Brain **17**:27, 1894.
36. Reimer: Jahrb. f. Kinderh. **10**:67, 1876.
37. Hada, B.: Virchows Arch. f. path. Anat. **214**:206, 1913.

Chronic infections, such as tuberculosis and syphilis, have been suggested as a cause of subdural membranes by Poumeau [38] and Heubner.[7e] In Poumeau's case the condition terminated fatally in miliary tuberculosis. Heubner studied an 18 week old child with congenital syphilis. He believed the infectious process produced an injury of a vessel, with bleeding into the dura, skin, muscles, and other parts.

Schuberg [3] and Kaplan [39] suggested alcoholism as the cause of vascular injury and subdural bleeding. Schuberg recorded 11 cases in persons suffering from alcoholism, without any history of associated trauma.

Ciarla [40] found 57 cases of subdural hematoma in 6,631 autopsies. In few of the cases did he observe a hematoma following trauma. Associated with the dural lesion in 20 cases was nephritis; in 13, a lesion of the brain, such as softening; in 7, alcoholism; in 6, syphilis; in 4, tuberculosis; in 4, malaria; in 2, trauma; in the rest, miscellaneous conditions. The author concluded that pachymeningitis haemorrhagica interna was related primarily to sclerosis of vessels, associated with age and accompanied by such conditions as alcoholism, chronic and acute infections and head trauma.

Experimental Investigations.—The experimental work so far reported seems to have shed little light on the causes of the subdural membrane. The reason will become obvious after discussion of the histologic character and pathogenesis of the lesion.

In 1868 Kremiansky,[5] strongly advocating alcoholism as the etiologic factor in the lesion, fed puppies each 1 ounce (31 cc.) of brandy daily for from two to five months and on killing them found slight hemorrhagic membranes on the inner surface of each dura. Sperling,[7b] using rabbits, introduced whole blood, defibrinated blood and a solution of iodine, hydrochloric acid and acetic acid into the subdural space. The whole blood after a period of three weeks became organized on the inner dural surface, producing a membrane of the type characteristic of pachymeningitis. Organization began after eight days. The defibrinated blood became resorbed, leaving no membrane. The other irritative solutions produced only mild thickening of the dura. Spiller and McCarthy,[41] using cats and dogs, nicked the superior longitudinal sinus sufficiently to produce a subdural hematoma. By putting an animal to death from time to time, they found that new membrane formation was distinct in five days and was fairly complete in five weeks. Marie,

38. Poumeau, M.: Bull. Soc. anat. de Paris **15**:60, 1841.

39. Kaplan, A.: Brain **54**:430, 1931.

40. Ciarla, E.: Arch. f. Psychiat. **52**:439, 1913.

41. Spiller, W. G., and McCarthy, D. J.: J. Nerv. & Ment. Dis. **26**:677, 1899

Roussy and Laroche [42] injected into rabbits and dogs subdurally pure blood, blood plus staphylococci and mild irritants, such as a mixture of fatty acid plus sodium nucleinate. Pure blood alone failed to produce a membrane in any of 9 animals. Infected blood and mild irritants resulted in membrane formation in a few animals and a hemorrhagic membrane in but 1. No histologic descriptions were given. The authors concluded that pachymeningitis haemorrhagica interna was not primarily a hemorrhage, because whole blood failed to produce such a membrane. Putnam and Putnam,[43] with 15 cats and 3 dogs, injected whole blood into the subdural space. Five animals showed no changes. In the rest within five days a thin neomembrane of fibrous tissue covered with mesothelium was found over the surface of the clot. The clot showed regressive changes; it was thin and traversed by connective tissue. Blood vessels were found only after the first week. Subsequent hemorrhages in the new membrane did not occur in any of their cases. Barret [44] in 1902 studied bacteriologically 23 cases of subdural hematoma, with negative results. In 8 instances, under aseptic precautions, he transplanted portions of subdural membrane into the subdural spaces of cats and dogs, with entirely negative results.

Apparently a true progressive chronic hematoma of the dura has as yet not been produced experimentally.

A review of 31 cases of subdural hematoma in which pathologic material was available for study revealed a most varied etiologic background. In only 6 was there presented a definite history of trauma. However, cases of subdural hematoma occurring in association with severe injury of the skull and brain were omitted. An attempt was made to limit the study to those cases in which the hematoma was the predominant finding and probably the only cause of death. In 3 cases the hematoma was associated with pernicious anemia and there was evidence of other bleeding besides the dural, such as scattered petechiae throughout the viscera. Alcoholism and syphilis were prominent among the 31 cases, occurring in 8. Two of the cases concerned aged persons and the lesions were assigned to sclerotic changes in the meningeal vessels with rupture. In a large group no etiologic factors were presented in the histories that might in any way account for the bleeding.

If one now considers the various factors that have been reported as entering into the causation of subdural hematomas, the following etiologic classification might be proposed:

---

42. Marie, P.; Roussy, G., and Laroche, G.: Compt. rend. Soc. de biol. **74:** 1303, 1913.

43. Putnam, T. J., and Putnam, I. K.: J. Nerv. & Ment. Dis. **65:**260, 1927.

44. Barret, J. O. W.: Brain **25:**181, 1902.

ETIOLOGIC CLASSIFICATION OF SUBDURAL HEMATOMAS

A. Hemorrhagic origin (new membrane formed on organization of the hematoma)

1. Spontaneous hemorrhage due to a primary vascular injury
   (*a*) Associated with hemorrhagic diathesis
      (1) Pernicious anemia
      (2) Scurvy
      (3) Purpura
      (4) Leukemia (vessel changes)
   (*b*) Associated with acute infectious diseases
      (1) Whooping cough
      (2) Measles
      (3) Diphtheria
      (4) Acute sepsis
   (*c*) Associated with chronic infectious diseases
      (1) Syphilis
      (2) Tuberculosis (vessel involvement)
   (*d*) Associated with alcoholism
   (*e*) Associated with degenerative conditions in blood vessels (arteriosclerosis)

2. Traumatic hemorrhage

B. Exudative origin (new membrane formed on organization of the inflammatory dural exudate, with bleeding occurring secondarily from the newly formed vessels)
   (*a*) Due to some undetermined infectious process of the dura
   (*b*) Associated with acute infectious diseases and due to the toxic or bacterial action on the dura
   (*c*) Secondary to a syphilitic involvement of the dura
   (*d*) Secondary to an involvement of the dura as a result of alcoholism and scurvy

SYMPTOMS

Since the symptoms ordinarily encountered in patients with subdural hematoma are protean, one cannot describe a typical picture in this condition. The symptomatic aspects may be variable in the extreme. Most often there is slow development of evidence of intracranial irritation—headache, vomiting and drowsiness, associated with mild or severe focal symptoms and definite changes in the patient's mental makeup. Occasionally the onset is abrupt, with sudden unconsciousness and without any prodromal complaints. The latter indicates a most grave type that rapidly terminates fatally if no intervention is attempted. The former characterizes types that are much less serious, in which distinct remissions are frequent.

Headache is perhaps the most common symptom and was the first complaint in over half the 31 cases. It may vary from a dull ache to a

severe pain. There is nothing localizing about the headache. It is at first periodic but later becomes more persistent and more intense. Nausea and vomiting may be associated with the headache. Projectile vomiting and severe vertigo were present in but 2 of the cases. Diplopia, although often present, is transient. Papilledema is common.

For the most part, repeated examination of the reflexes does not help to localize the lesion, the reflexes often varying from day to day. Ultimately the patient may show a definite irregularity of both superficial and deep reflexes which indicates the side of the lesion. There may also occur gradual paresis of the limbs, which is usually more marked on one side. Such patients have difficulty in walking, dragging the involved limb. There may eventually result complete paralysis. In 6 of the 31 cases studied there were definite and persistent unilateral localizing findings. In 4 there were diffuse and variable reflex changes.

In some cases the mental picture predominates. The patients become morose, irritable, clouded, untidy and unmanageable. There is definite clouding of the sensorium, the patients often appearing stupid and bewildered. Intelligence and memory are impaired, and there results distinct mental retardation.

Consciousness may be lost either gradually or suddenly. This loss of consciousness may be preceded by convulsions or delirium.

The clinical picture may vary from morning to evening and indeed throughout the entire duration of the illness, so that no localization can be made with any degree of certainty. Frequently one finds a lesion on the same side as pyramidal signs, due to a pushing by the hematoma of the opposite side of the brain against the cranial vault. Often the observations at autopsy are out of all proportion to the scanty and varying neurologic signs. This variability results in uncertainty in diagnosis and often causes too great a delay in diagnosis and therapy.

The spinal fluid, as a rule, is normal, since a hemorrhage strictly confined to the subdural space will obviously not stain the spinal fluid. Occasionally xanthochromic fluid is obtained, and sometimes the fluid varies from a blood-tinged color to a deep yellow or straw color. In only 2 of the cases reviewed here were there red cells in the fluid.

The picture in infants is quite different from that in adults. The ability of the sutures in childhood to give way accounts for this difference. The symptoms may be gradual or sudden in onset and are often those of increased intracranial pressure with meningeal irritation. The first symptom is frequently gradual enlargement of the head, usually associated with vomiting, convulsions and cervical rigidity. Alteration of the reflexes, muscle twitching, paralysis, hyperesthesia and somnolence may occur. In children, retinal hemorrhages occur in about 50 per cent of cases but are not pathognomonic of hemorrhagic pachymeningitis, as they may be seen in association with tumors, menin-

gitis and primary hydrocephalus. Rosenberg [14] described three courses which this condition has been observed to follow in children: 1. A slow course with no signs of involvement of the nervous system, only restlessness and gradual enlargement of the head, with widening of the fontanel. 2. An acute course with rapid onset of symptoms, beginning with vomiting, restlessness and crying. In the patients observed the course varied in duration but eventually resulted in spasticity of extremities, increased reflexes, stiff neck and somnolence. The head enlarged rapidly and convulsions were frequent. 3. A fulminating, rapidly fatal form, with symptoms of acute meningitis and rapid involvement of the entire nervous system. The patients became comatose with high fever and stiff neck.

The diagnosis is established on puncture of the fontanel. In the case of hematoma, just after piercing the membranous fontanel a straw-colored to almost pure bloody fluid is obtained.

The mortality is great in infants even in cases in which operation has been attempted. Repeated drainage by way of the fontanel is often the selected treatment. Many of the infants who live through attacks carry a permanent disability such as chronic hydrocephalus, imbecility, blindness, deafness, paralysis, spasticity or speech defect. Rosenberg found that 70 per cent of his recovered infants had residual defects.

MORBID ANATOMY

Gross Appearances.—The usual location of the subdural hemorrhage is in the frontal and parietal regions, although a large hemorrhage may cover an entire hemisphere. The hemorrhage is bilateral in about one third of the reported cases.

The hematoma may vary in size from petechial and ecchymotic formations to massive clots which displace a large part of a cerebral hemisphere. The small hemorrhages are undoubtedly far more common than the large. They produce few or no symptoms and are usually overlooked or ignored in routine postmortem study. They frequently undergo recessive changes and absorption, leaving either a rusty membrane on the inner surface of the dura or a few areas of residual hemosiderin granules.

In a routine study of the pachymeninges from patients dead of various severe toxic conditions, many hemorrhagic areas can be found within the inner dural layer. In some cases these extravasations have reached a quite formidable size, while in others they consist merely of patchy discolored areas made up of residual hemosiderin granules, the remains of a previous bleeding. These findings represent the very earliest stages in the formation of subdural hematomas. Depending on the degree of the vascular injury, the process may stop at this early stage or progress to a more severe involvement. The first stages in the

pathogenesis of subdural membranes might well be studied by an investigation of these very early lesions.

In more extensive involvement, the dura presents a much more varied appearance. In some cases it becomes thickened and shows brownish discoloration within its inner surface. In other cases a definite hematoma may be present, composed either of fluid or of clotted dark brown or black blood. Usually the younger lesion contains liquid blood with only beginning coagulation. The older hemorrhage is completely coagulated. It appears dark brown or black, thickest in the center and very thin at the periphery, where it blends with uninvolved dura. An area of discoloration can be found surrounding the hematoma. The involvement may be diffuse, forming a fine layer over the entire inner

TABLE 1.—*Bilaterality of Lesion in Cases of Subdural Hematoma*

Author	Total Number of Cases	Number in Which Hemorrhage Was Bilateral	Number in Which Hemorrhage Was Unilateral	Age Group
1. Schuberg [3]	25	14	11	Adults
2. Ciarla [40]	44	13	31	Adults
3. Rand, C. W.: Arch. Surg. **14**: 1136, 1927	7	0	7	Adults
4. Sherwood [33]	9	6	3	Children
5. Kaplan [29]	8	1	7	Adults
6. Fleming and Jones [46]	8	4	4	Adults
7. Leary [30]	50	7	43	Adults
8. Grant, F. C.: J. A. M. A. **105**: 845, 1935	16	3	13	Adults
9. Frazier [25]	6	3	3	Adults
10. Baker (present report)	31	10	21	Mixed
Total	204	61	143	

surface of the pachymeninx, or may be localized to certain isolated regions. In most instances the hematoma is covered by a fine transparent membrane.

The most striking type of lesion and the one most likely to produce symptoms consists of a large collection of fluid or coagulated blood, involving an extensive area of the dura and enclosed within a fairly heavy connective tissue covering. The content varies a great deal depending on the age of the lesion. At first it consists of either liquid blood or of a black gelatinous clot resembling that found in any hemorrhage. As the coagulum becomes encapsulated, its content softens and liquefies to form a coffee-colored fluid in which float fragments of tarry or brownish-red disorganizing clots of varying size. Still later the fluid becomes greenish or yellow but still contains tarry blood, partly fluid and partly clotted. A greenish discoloration beneath the dura is quite characteristic of this condition. The yellow color of the underlying arachnoid and pia is assumed to be due to transformation of hemoglobin into biliverdin and bilirubin. Many large hematomas never

liquefy but remain as thick layers of black clotted blood compressing the underlying brain tissue.

The enclosing membrane is variable in appearance. In some cases it is composed of a very fine, delicate, transparent layer that is adherent to the dura only at the periphery of the hematoma. In other lesions, this membrane is thicker and less transparent, forming a fairly tough opaque gelatinous or fibrous layer. The dural surface of the membrane is loosely adherent to the hematoma, and when stripped away it appears rough and coarse and contains many small particles of adherent clot. Internally, on the side toward the arachnoid, the membrane is smooth and shiny. It is not adherent to the arachnoid. Occasionally the membrane wall contains cartilage and even bone.

The subdural hematoma is usually flat. In the more extensive forms it produces a definite compression of the brain. The deformity is caused by pressure from the slowly forming hematoma, which displaces the brain tissue, making a bed for itself. The underlying convolutions are flattened, the cerebral hemisphere on the side of the hematoma being compressed sometimes to one-half its volume. The brain is most often displaced downward and inward, with resultant distortion and narrowing of both ventricles. Continued pressure produces atrophy of the underlying cerebral cortex.

Microscopic Appearances.—The microscopic observations are based on a study of pachymeninges in 61 cases. Of these duras, 17 were from apparently normal persons, while 13 were from patients dead of severe toxic conditions. The latter duras contained small extravasations, and study of these enabled one to follow the formation and fate of the smaller and less conspicuous hemorrhages. The rest of the material was composed of 31 typical subdural hematomas.

In each case sections were taken from various regions of the apparently normal and the involved portions of the dura. These were stained with the hematoxylin-eosin stain, by the Mallory-Heidenhain [44a] technic (azocarmine), with Weigert's elastic tissue stain and with Bielschowsky's silver stain.

(*a*) Normal dura. The dura forms the outer boundary of the subdural space. It is a tough fibrous membrane which has a rough external and a smooth glistening internal surface. The subdural space is a potential space between the dura and the arachnoid and contains only a slight amount of fluid. The cranial blood sinuses are found within the dura.

The pachymeninx is composed roughly of two indefinite layers of dense fibrous tissue, which are inseparable for the greater part of their extent. The bundles of connective tissue intertwine and cross one another rather obliquely and are frequently separated by grooves or

44a. Kernohan, J. W.: Am. J. Clin. Path. **4**:421, 1934.

interspaces of various shapes and sizes. The midportion of the dura usually shows the largest number of these round and oval spaces, the so-called lacunae. The lacunae are, as a rule, empty and contain no blood, pigment or fibrin. The inner surface of the dura is often covered by a single layer of cuboid cells. The cell body is difficult to outline, but the nucleus appears large and vesicular, with a very fine chromatin pattern and a very thin nuclear membrane. This epithelial or mesothelial layer is usually not complete and may lie immediately on the dense fibrous dura, or, as is more frequently the case, it may be separated from the fibrous dura by a thin layer of loose connective tissue called the subepithelial layer.

Blood vessels, primarily capillaries, are very few within the dura and are hard to make out except when vascular congestion has occurred. When visible, these vessels, although mere capillaries, are remarkably large, many of them in their normal condition having a diameter of from three to four times that of ordinary capillaries. They lie in grooves that run chiefly parallel with the connective tissue fibers and appear as huge blood-filled spaces, each with a most delicate wall composed of a single layer of elongated cells. Most of the dural vessels are situated in two positions: A layer of vessels can be found in the midportion of the dura, between the two inseparable layers of collagenous fibers. The richest capillary network, however, is found in the delicate loose connective tissue that lines the innermost portion of the dura, the subepithelial layer.

The dura, therefore, from within outward contains the following layers:

1. Epithelial or mesothelial lining
2. Subepithelial layer of loose connective tissue
3. Inner capillary network—contained in layer 2
4. Inner fibrous tissue layer
5. Outer capillary network
6. Outer dense fibrous tissue layer

To study the pathogenesis of subdural hematoma, it becomes necessary to investigate all degrees of bleeding from very mild to massive hemorrhage, the latter resulting in the typical well known hemorrhagic membrane. Subdural bleeding of varying degrees is frequently produced in severe toxic states, and a study of the fate of extravasations in such states proves helpful in completing one's knowledge concerning the finer details in the development of the subdural membrane.

(*b*) Mild bleeding. Hemorrhage, when it occurs, is found either between the fibrous layers of the dura or on the inner dural surface, i. e., in the region of the outer or in that of the inner capillary network.

In either location a mild hemorrhagic process does not tear or injure the surrounding tissue, but the erythrocytes remain localized to the dura, undergo hemolysis and produce a mild reaction of the surrounding tissue. The latter consists in a proliferation of fibroblasts with production of many spindle-shaped cells and new collagenous fibers. Unless more extensive hemorrhage occurs, such a mild process ends in the deposition within the dura of more strands of collagen fibers. Within the new cells or between the dural fibers are numerous granules of hemosiderin; these produce a patchy or diffuse brownish discoloration to the dura.

(*c*) Moderately severe bleeding. Hemorrhage of a moderate degree produces variable results. Within the fibrous layers of the dura such hemorrhage is usually limited and localized. When extensive, it tends to split the layers of the dura; the inner lamina then forms the fibrous inner covering of the hematoma (fig. 1 *A*). In the older person the dural tissue seems more brittle, and bleeding often produces fragmentation of the fibrous elements, with the red cells tearing through the ruptured fragments and thus reaching the looser connective tissue of the subepithelial region, which is then in turn torn away to produce the partial limiting membrane.

Most often the larger hemorrhage occurs in the region of the inner capillary bed and dissects along the loose connective tissue of the subepithelial layer, portions of which usually remain as a partial membrane to the subdural hematoma. It is these fine fragments of connective tissue that remain scattered throughout the hematoma and play such an important part in its localization. It seems, therefore, that in most cases of subdural hemorrhage a portion of the original pachymeninx is split off and acts as an inner limiting membrane to the coagulum. This accounts for the presence of such a limiting tissue even in the fresher hematoma in which the formation of new connective tissue could not possibly have occurred.

The presence of the blood clot induces certain reactions on the part of the dura and results in the penetration into the clot of dural vessels, accompanied by fibroblasts (fig. 1 *B*). The red cells break down into a granular débris. The coagulum begins to be penetrated by new capillaries which grow out from the dura and are accompanied by connective tissue cells. This penetration may occur along the entire surface of the coagulum or, as is seen more frequently, only in certain localized regions. The organization progresses slowly to the surface of the hematoma, producing the false membrane. During the early stages the ground substance of the clot would consist of numerous small and large vessels, many fibroblasts, various blood extravasations and pigmented areas, hemolyzed red cells and some macrophages. The spindle-shaped fibro-

blasts are by far the most active elements. They proliferate rapidly and fill the tissue between the new vessels, eventually compressing and obliterating all the adjacent elements. They lay down many fine and

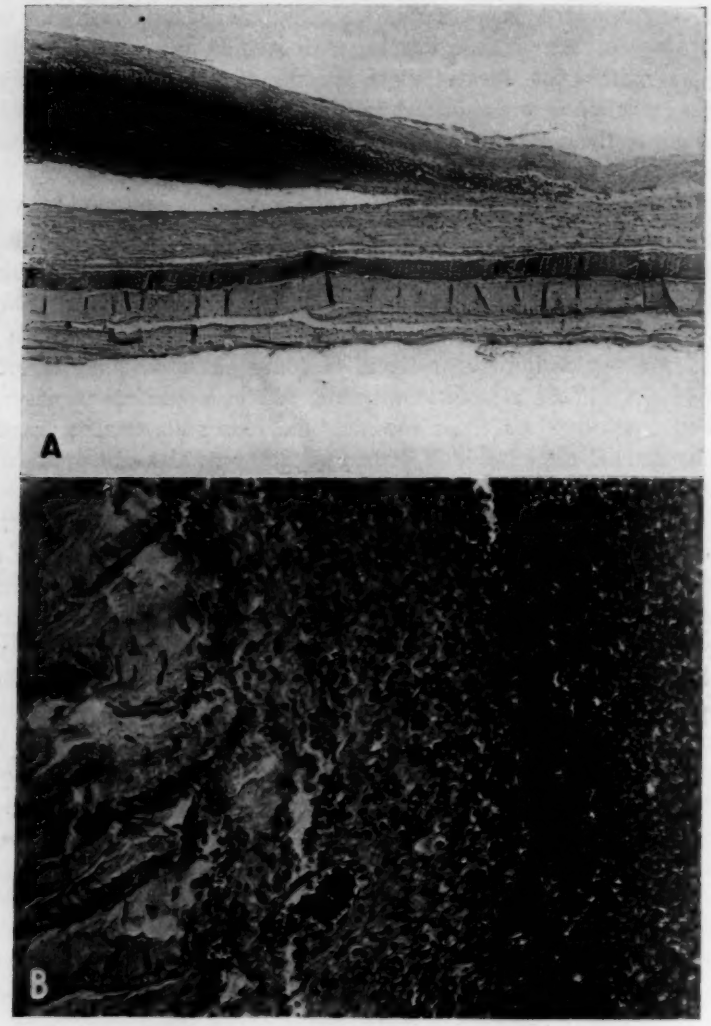


Fig. 1.—*A*, a section through a moderately severe intradural hemorrhage. Note how the dura is being split by the bleeding, the inner dural tissue forming a cover for the hematoma. *B*, early organization of a dural hematoma. The capillaries and fibroblasts can be observed invading the clot from the dura.

coarse strands of collagen, many of which seem to merge with the underlying dura and form the basis of the adherence of the new membrane to the pachymeninx (fig. 2 *A*). Since the fibroblastic proliferation starts

at the dural region, this portion of the new membrane is the first to become organized and already appears fibrous while the inner layers of the clot are just beginning to be penetrated by the invading cells and capillaries (fig. 2*A*). Blood pigment can be observed filling the macrophages and fibroblasts. The latter are frequently engorged with these brownish granules, which extend outward even into the bipolar ramifications of the cell processes. Pigment may also be seen scattered diffusely between the fibroblasts and collagenous fibers.

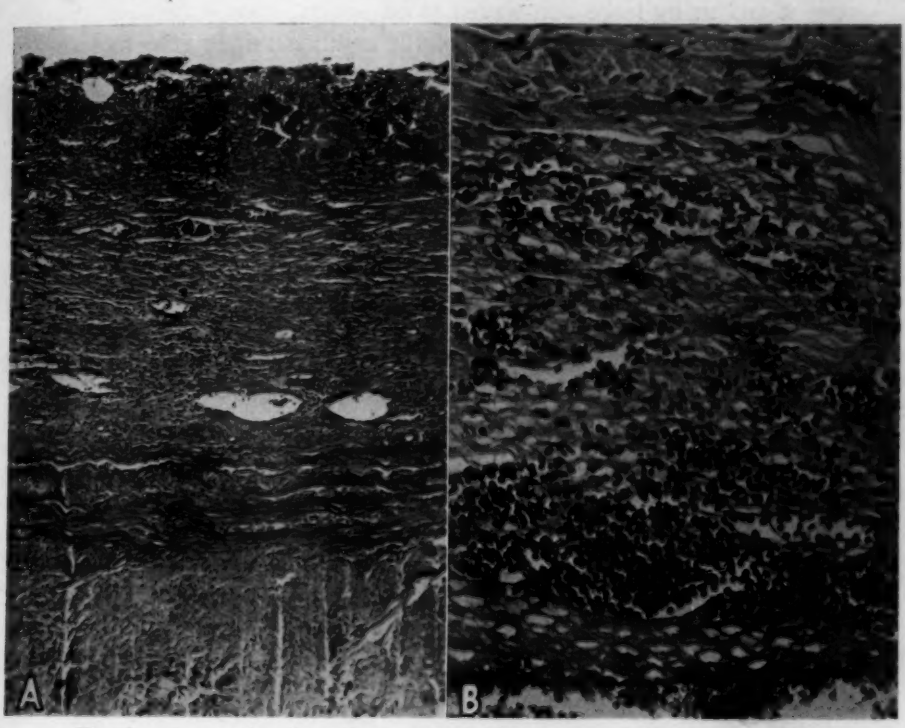


Fig. 2.—*A*, a section through a pseudomembrane formed as the result of almost complete organization of a dural hematoma. The new membrane is most fibrous in that region which lies adjacent to the dura. Its inner, or arachnoid, portion is as yet not organized. There are numerous large blood spaces scattered throughout the new connective tissue. *B*, a section through a completely organized subdural hematoma. In this case the blood spaces are much distended and show an early secondary bleeding into the newly formed membrane.

After the deposition of the fine collagenous strands there are formed between them numerous large clear spaces that are either empty or contain a few red cells. These spaces are really blood channels and on close examination are seen to be lined by a very fine layer of cells having elongated, flattened hyperchromatic nuclei. The cell bodies are extremely difficult to make out, but occasionally streams of protoplasmic

processes can be detected between their nuclei. These blood spaces are often congested and show tiny tears and extravasations of red cells; no doubt they are the source of the secondary hemorrhages into the newly formed membrane (fig. 2 *A* and *B*).

At this stage in the process of organization, the newly formed membrane has a fairly characteristic appearance. That portion of the membrane adjacent to the dura is composed of fairly loose connective tissue containing many active proliferating fibroblasts filled with blood pigment and many large thin-walled blood spaces situated between the fibrous strands. The inner portion of the new membrane (the surface toward the arachnoid) shows only slight invasion by fibroblasts and capillaries and still contains many hemolyzed red cells, macrophages and blood pigment. The inner surface of the membrane is covered by a capsule derived from the dura at the time of the rupture.

In the later stages of organization the entire clot undergoes complete replacement by fibrous tissue. The dural portion of the new membrane becomes very fibrous and is composed of large bands of coarse collagen fibers containing but few cells or vessels, while the more recently organized inner or adventitial portion of the membrane consists of loose connective tissue elements with many fibroblasts and huge blood spaces. Frequently the new membrane merges with the dura and is hard to separate even microscopically. As a rule, however, the connection remains quite distinct and, peculiarly enough, quite fragile, only a few fibrous strands spanning the two structures.

If no secondary hemorrhage results within the subdural membrane, the entire new structure undergoes complete fibrosis. The capsule or covering is usually composed of a somewhat denser tissue, but in none of the cases studied was I able to detect an epithelial or mesothelial lining on the inner surface of the membrane.

In massive subdural hematoma, where the erythrocytes have torn through the surface and have formed a coagulum many times thicker than the dura itself, the process of organization is somewhat different. The capillaries and accompanying fibroblasts grow out from the dura into the clot, but the invasion occurs from localized foci and involves only scattered areas of the hematoma. From these separated areas of proliferation the vessels and fibroblasts grow rapidly through the clot to its innermost surface, resulting in the production of long strands of fibrous and granulation tissue that extend from the dura to the surface of the clot and divide the hematoma into small sections. The fibroblasts that reach the inner surface of the hematoma proliferate and produce a fine limiting membrane. The proliferating tissue within the coagulum produces small projections at right angles to the original growth of fibroblasts. These outshoots proliferate rapidly and join

with those from adjacent strands, with ultimate complete replacement of the clot by the process of organization. Naturally, the most mature fibrous tissue will be found within the long strands that pass directly through the hematoma, since these were the first deposited.

(*d*) Signs of inflammation. A feature not infrequently seen within the newly formed membrane is the presence of many cells of an inflammatory nature. They are mostly small mononuclears, intermixed with a few polymorphonuclears, and may be found within localized areas or may be seen scattered diffusely throughout the dural region of the new membrane. The membrane containing these inflammatory cells as a rule possesses but few hemorrhages and rarely shows old blood pigment within its granulation tissue. The appearance is suggestive of a primary inflammatory process with membrane formation, and the mild hemorrhages seem to be secondary from the newly formed, thin-walled vessels.

(*e*) Secondary hemorrhage within the new membrane. If the factors responsible for the original bleeding are not removed, there occurs within the new membrane a congestion of the large, thin-walled blood spaces (fig. *2B*). This is a common appearance. Slight rupture and extravasation of red cells soon produce scattered areas of erythrocytes. If the new membrane has undergone fairly complete fibrosis, the heavy collagenous bands localize the bleeding even though it is fairly severe. Usually, however, the secondary hemorrhage occurs fairly early in the formation of the new membrane. The erythrocytes dissect between the loose fibers and tear the still frail tissues to reach the inner surface of the membrane. The red cells tend to follow the pathway of least resistance, which is invariably toward the inner, or arachnoid, surface of the new membrane, since it is this region that is composed of the newest and hence the weakest connective tissue strands. This region also does not have the support of the underlying heavy fibrous dura. Secondary bleeding may, therefore, tear through the inner portion of the new membrane to produce a new surface bleeding or, as is more common, may split the new membrane by dissecting longitudinally between the interstices of its fibers (fig. *2B*). In the latter case the innermost layer of the new membrane would comprise the limiting tissue of the new hematoma. Since this portion of the membrane is usually composed of young fibroblasts, these cells continue to mature and produce collagen, a process which results in the formation of a heavy limiting layer for the new bleeding. Organization of this new clot will occur in the manner already discussed.

Jores [45] believed it possible to differentiate the membrane in traumatic from that in idiopathic pachymeningitis haemorrhagica interna.

45. Jores: Verhandl. d. deutsch. path. Gesellsch. **1**:49, 1899.

He postulated that the former consists of connective tissue rich in fibroblasts with only small, thin-walled, slowly growing vessels. In true pachymeningitis, the membrane consists of homogeneous connective tissue with fewer nuclei but filled with large, actively growing vessels.

Putnam and Cushing,[27] duplicating the work of Jores, also believed that the nontraumatic hematoma of the dura is histologically distinguishable from the traumatic. The nontraumatic, idiopathic, vascular type of membrane is composed of a network of large blood vessels which have the structure of capillaries although they reach a diameter of 40 microns. The traumatic membranes are characterized histologically by the presence of large irregular blood-filled spaces much larger than the "giant capillaries" earlier described. These spaces lie parallel with the dura and are separated from it by a few fibroblasts. These spaces are mesothelial lined and anastomose with each other and with the apparently normal capillaries within the membrane.

Of the 31 cases of subdural hematoma reviewed here, 6 were traumatic. Microscopically, however, the process of organization produced a similar histologic picture in all membranes. This is not surprising, since a coagulum once formed, regardless of the cause, produces certain definite tissue reactions and a fairly uniform histologic picture.

TREATMENT

At present most authors are agreed on the treatment of subdural hematoma. Once such a lesion is suspected, four trephine openings are made, under local anesthesia, in the frontoparietal and parieto-occipital region of each side of the head. The incisions are so fashioned and the openings so placed that they may be incorporated in an osteoplastic flap at a subsequent date if necessary. If a hematoma is discovered, it is exposed through small openings in the dura. Fleming and Jones [46] advised irrigation of the sac with Ringer's solution and closure of the incision without drainage. Kaplan [39] believed that as much of the membrane as will come through the trephine opening should be removed. He maintained that the condition rarely recurs. As a rule the patient regains consciousness shortly after removal of the clot. Failure to react promptly means that the patient has either an additional cerebral laceration or a hematoma on the other side. Munro [47] believed in decompression for the subdural hematoma. He removed the entire clot and provided drainage. Bowder [48] used the encephalogram instead of buttons as aids in the diagnosis of the subdural hematoma. He injected from 50 to 60 cc. of air with less aftermath than is entailed in an exploratory

46. Fleming, H. W., and Jones, O. W.: Surg., Gynec. & Obst. **54**:81, 1932.
47. Munro, D.: J. Nerv. & Ment. Dis. **80**:312, 1934.
48. Bowder, E. J.: J. Nerv. & Ment. Dis. **80**:313, 1934.

decompression. He admitted that it may seem radical to introduce air in the presence of a laceration of the brain, but so far no untoward results have been observed.

In children the treatment of the subdural hematoma has been unsatisfactory. The mortality has been high in those treated radically and the sequelae frequent in those treated conservatively.

TABLE 2.—*Summaries of Cases of Subdural Hematoma*

Case	Age of Patient, Yr.	Sex	Position of Lesion	Probable Cause	Duration
1	48	F	Bilateral	Pernicious anemia	7 days
2	47	F	Unilateral	Pernicious anemia	?
3	42	F	Bilateral	Pernicious anemia	60 days
4	70	M	Bilateral	Syphilis	24 days
5	54	M	Unilateral	Alcoholism	?
6	41	M	Unilateral	Alcoholism	8 days
7	56	M	Unilateral	Alcoholism	3 days
8	55	M	Unilateral	Atherosclerosis	20 days
9	76	M	Bilateral	Marked atherosclerosis	37 days
10	38	M	Unilateral	Trauma	?
11	45	M	Unilateral	Trauma	3 days
12	33	M	Bilateral	Trauma	59 days
13	22	M	Unilateral	Trauma (?)	?
14	70	M	Unilateral	Trauma	42 days
15	68	M	Unilateral	Thrombosis of superior sagittal sinus	2 days
16	25	F	Bilateral	Trauma	8 days
17	34	M	Unilateral	Syphilis	1 year? Few hours?
18	29	M	Unilateral	Syphilis	8 days
19	29	F	Unilateral	Chronic alcoholism Syphilitic meningitis	4 days
20	38	F	Unilateral	Chronic alcoholism	7 days
21	55	F	Unilateral	Metastatic carcinoma to dura from breast	?
22	53	M	Bilateral	None	50 days
23	7 weeks	F	Bilateral	None	?
24	33	M	Bilateral	None	60 days
25	61	M	Unilateral	None	32 days
26	54	M	Unilateral	None	7 mo.
27	46	M	Unilateral	None	40 days
28	65	F	Unilateral	None	5 days
29	49	M	Unilateral	None	7 mo.
30	50	M	Bilateral	None	4 days
31	30	M	Unilateral	None	11 days

COMMENT

A review of the literature and of the pathologic material available at the University of Minnesota suggests certain facts concerning the subdural hematoma that warrant emphasis. It appears certain that the etiologic background of the so-called subdural hematoma is quite variable and is not limited to any single condition. In many cases trauma plays an important role, but one must be guarded in insisting that hematoma is in every case due to trauma and then in many cases accepting a trivial injury as important.

When the hematoma is traumatic, the source of the bleeding, on gross inspection, often appears to be a torn cerebral vein or sinus. However, from the histologic investigations reported here one can certainly question such a source as the only or even the primary one for the hemorrhage. If the hematoma arises from a ruptured vein or sinus, as has been asserted by many investigators, it becomes difficult to explain the presence of a limiting membrane within a few days after the injury. A much longer time is required for the production of such a membrane. Moreover, since the source of this limiting tissue must be the dura itself, it appears unlikely that the invading fibroblasts and capillaries would wander through the entire hematoma to form a membrane on its inner, or arachnoid, surface rather than produce a gradual and progressive organization of the entire clot, beginning at the inner surface of the dura. Since limiting membranes are frequently present very early, even before organization has begun, it is more logical to assume that the original trauma caused not only an injury to the veins or sinuses (if such did occur) but also a rupture of the intradural vessels, producing an intradural hematoma. The inner fibrous tissues of the dura would then be torn away by the hemorrhage and would form the limiting membrane. Histologic investigation produces overwhelming evidence in favor of the view that subdural hematoma, regardless of its etiology, is in reality intradural hemorrhage. Although the term "subdural" has been widely accepted and used, it is an incorrect description of the process that has occurred.

It may be this misconception of the pathogenesis of the hematoma that accounts for the varied results reported from experimental investigations. All investigators have attempted to produce the hemorrhagic dural membrane by injecting substances into the subdural space. To duplicate the formation of a subdural hematoma, it is obvious that the various substances ought to be introduced into the layers of the dura. If this technic were adopted, it is possible that the results obtained would be more uniform. Boekmann's [32] observations certainly lend support to this view. In each of a number of cases, as a result of some surgical procedure, he actually saw a layer of blood form on the inner surface of the dura. Yet, at autopsy, in none of these cases were there any signs of a hemorrhagic membrane, the blood having been completely absorbed.

The traumatic membrane is usually composed of a single layer. This is to be expected since the trauma which is the causative factor is as a rule not repeated. On the other hand, the membrane resulting from the bleeding of a vessel secondary to a hemorrhagic diathesis or secondary to an acute or a chronic infection is frequently composed of numerous layers, since in many of the cases the causative factor continues to be active.

In certain cases the hemorrhagic membrane is produced by a purely exudative process within the dura. During recent years even this has been classed as subdural hematoma. It would seem much more appropriate to identify such a process by the name originally applied to it by Virchow, i. e., "pachymeningitis haemorrhagica interna," since the lesion really represents a true inflammation of the dura.

CONCLUSIONS

The etiologic character of the subdural hematoma is quite variable. In a few cases it arises from an exudative process within the dura and may very well be called pachymeningitis. Trauma also plays an important role, but is by no means the only other factor. Hematoma as a result of bleeding from dural vessels may occur in the hemorrhagic diathesis, in acute and chronic infectious diseases, in chronic alcoholism and in terminal degenerative conditions, as atherosclerosis.

In most cases the subdural hematoma is really an intradural bleeding. Hemorrhage occurs from the two layers of dural capillaries and splits off a portion of the original pachymeninx, which acts as the inner limiting membrane to the coagulum.

Notes and News

University News, Promotions, Resignations, Appointments, Deaths, etc.—Albert Casey, of the department of pathology at St. Louis University, has been appointed assistant professor of pathology and bacteriology at the Louisiana State University, New Orleans.

William M. Hale, of the department of bacteriology at Yale University, has been appointed professor of bacteriology at the college of medicine of the State University of Iowa.

A. E. Boycott, professor emeritus of pathology at the University of London, has died at the age of 61 years. He was at one time editor of the *Journal of Pathology and Bacteriology*.

Vannevar Bush, vice president and dean of engineering of the Massachusetts Institute of Technology, has been elected president of the Carnegie Corporation of Washington. He succeeds John C. Merriam, who becomes president emeritus.

Maurice C. Hall, chief of the division of zoology of the National Institute of Health, of the United States Public Health Service, died May 1, at the age of 57 years.

William S. Tillett, professor of bacteriology at New York University College of Medicine, has been appointed professor of medicine at that institution.

Serodiagnostic Tests for Syphilis.—A meeting to consider problems connected with serodiagnostic tests for syphilis will be held at Hot Springs National Park, Ark., Oct. 21-22, 1938. Laboratory workers, physicians and health officers interested in the control of syphilis are invited to this meeting. Those who wish detailed information should write to the Surgeon General, United States Public Health Service, Washington, D. C.

Society News.—The American Association of Pathologists and Bacteriologists at the recent meeting held at Atlantic City, N. J., elected the following officers for the ensuing year: president, Earl B. McKinley; vice president, Carl V. Weller; treasurer, F. B. Mallory; secretary, Howard T. Karsner. The association will meet next year on April 6 and 7 at the Medical College of Virginia, Richmond.

Abstracts from Current Literature

To Save Space the Original Titles of Abstracted Articles Sometimes Are Shortened

Experimental Pathology and Pathologic Physiology

The Response of Guinea Pig Bone Marrow to Liver Extract. E. A. Gall, Am. J. Path. **13**:575, 1937.

Five groups of guinea pigs were studied for the purpose of observing the effect of liver extract on the bone marrow and determining the mechanism of its action. Ten animals fed a diet of oats, carrots and lettuce showed densely cellular marrow with evidence of depressed maturation of its elements. Seven guinea pigs fed the same diet and given subsequently single doses of liver extract exhibited marked depletion of cellular content with abruptly accelerated maturation of the marrow elements. Two guinea pigs fed a similar diet and subsequently given daily doses of liver extract for six and ten days, respectively, manifested a return of cellularity, with persistence, however, of maturation. Twenty guinea pigs from the ordinary laboratory stock fed only hay, oats and carrots possessed a relatively densely cellular marrow which showed no inhibition of maturation. Small and varying numbers of megaloblasts were identified within the marrows of all five groups. The diet of oats, carrots and lettuce obviously lacked a substance available in both liver extract and a diet of hay, oats and carrots. This substance permitted maturation of immature hemic elements. From Author's Summary.

Experimental Coronary Occlusion. L. Gross, M. Mendlowitz and G. Schauer, Am. Heart J. **13**:647 and 664, 1937; **14**:21, 1937.

Experiments with Chest Open.—Determinations were made of pulse rate, venous blood pressure, hemoglobin percentage, erythrocyte count, blood volume and serum protein concentration first in anesthetized dogs and then in anesthetized dogs in which thoracotomy and dissection of the anterior descending branch of the left coronary artery without ligation was performed and finally in dogs in which, in addition to this procedure, the anterior descending branch of the left coronary artery was ligated. The essential resulting difference among the groups was found in the marked drop in the cardiac output of the third group. The drop was considerably greater in these animals in which the artery was ligated than in the two control groups. The preoperative level was reached or even surpassed within twenty-four hours.

Experiments with Chest Closed.—A technic was developed permitting the ligation of the coronary artery by means of a specially constructed knot a week or two after the wound in the chest was closed. This permitted elimination of the thoracotomy factor at the time of ligation. The findings were essentially the same as in the first group of experiments—diminished cardiac output and left ventricular congestive failure. No satisfactory explanation is offered for the diminished cardiac output.

Experiments with Heart Denervated.—To test the influence of neurogenic factors a denervation of the heart was undertaken. It involved resection of both thoracic sympathetic chains, as well as bilateral sympatheticovagotomy in the neck. The only possible remaining nervous pathway was through the carotid sinus, which was left intact. The ligation of the anterior descending branch of the left coronary artery in dogs with denervated hearts was followed by a fall in the cardiac output and blood pressure. The result points against a neurogenic

origin of the decrease of cardiac output and in favor of its cardiogenic nature. The functional results of the ligation were cardiogenic cardiac failure and left ventricular congestive failure.

I. DAVIDSOHN.

HYPOTHALAMIC REGULATION OF TEMPERATURE IN THE MONKEY. S. W. RANSON, C. FISHER and W. R. INGRAM, Arch. Neurol. & Psychiat. **38**:445, 1937.

Lesions were produced in the hypothalami of monkeys (Macacus rhesus) by means of the Horsley-Clarke apparatus, and a study was made of the effect of such lesions on the regulation of temperature. When the monkeys with hypothalamic lesions were kept in a moderately warm temperature, they exhibited impaired regulation of heat—prolonged hyperthermia or hypothermia. When they were exposed to moderately low external temperatures, they lost heat rapidly, but when they were exposed to moderately high external temperatures, the rectal temperatures rose rapidly, as in a normal monkey. Only when the external temperature (in the boxes) was gradually increasing did the animals with hypothalamic lesions exhibit less resistance than the normal animals—the rise in the temperature was steady and continued throughout the test. The temperature rose to 106 F., while in normal animals it did not go higher than 103 F. Hyperthermia was transient, lasting about twenty-four hours; hypothermia lasted much longer, about a month. The areas affected in monkeys with hypothermia were dorsal and lateral to the rostral end of the mamillary body and were situated mainly in the lateral portion of the hypothalamus. In monkeys in which the lesions did not reach back to the mamillary bodies, hyperthermia was the result.

GEORGE B. HASSIN.

DOES LIVER SUPPLY FACTORS IN ADDITION TO IRON AND COPPER FOR THE REGENERATION OF THE HEMOGLOBIN IN NUTRITIONAL ANEMIA? E. B. HART, C. A. ELVEHJEM and G. O. KOHLER, J. Exper. Med. **66**:145, 1937.

The data indicate that the effectiveness of whole liver in the treatment of nutritional anemia induced in rats by a milk diet is directly proportional to its available iron and copper content. The other constituents in liver are not needed for maximal regeneration of hemoglobin on a diet of milk, iron, copper and manganese. Commercial preparations of liver products with iron or with iron and copper vary greatly in their hemoglobin-regenerating efficiency in rats with nutritional anemia. The variation is correlated directly with the iron and copper content of the preparation. When the copper-iron ratio was too wide, the regeneration of hemoglobin was checked, although the iron supply was sufficient for optimal regeneration.

FROM AUTHORS' SUMMARY.

TRANSPLANTS OF THE PROSTATE AND SEMINAL VESICLE. R. A. MOORE and others, J. Exper. Med. **66**:273, 281 and 291, 1937.

A method is described with which the two dimensional size of a transplant in the anterior chamber of the eye may be measured daily with a high degree of accuracy. The size of transplants of the prostate and seminal vesicles varies from day to day, and objective evidence indicates that this variation is related to physiologically active testicular tissue.

With a photographic method for the determination of the size of a prostatic or a vesicular transplant in the anterior chamber of the eye, it has been possible to follow continuously the response to an injection of a glandular preparation. The results may be briefly summarized as follows: (*a*) One injection of the gonadotropic substance found in the urine of pregnant women produces a moderate increase in size; (*b*) subsequent injections of this same substance for a period of at least three months are without effect; (*c*) an alkaline extract of the whole anterior lobe of the pituitary gland produces a similar increase; (*d*) all pituitary derivatives are ineffective in the castrated animal; (*e*) castration brings about a

decrease in size that gradually loses velocity; (*f*) androgen produces a slight increase in intact, and a variable, at times conspicuous, increase in castrated, animals; (*g*) estrogen provokes a conspicuous increase in both intact and castrated animals; (*h*) the hormone of the corpus luteum has no effect, and (*i*) there is no evidence of synergism of the pituitary factors and androgen nor of antagonism between androgen and estrogen in adult rabbits.

In male rabbits, injections of estrogen result in marked increase in the size of ocular transplants of the prostate and seminal vesicles and correlated elevation of the blood cholesterol. Other hormones which cause an increase in the size of the transplants do not produce the correlated rise in blood cholesterol. The ingestion of cholesterol, which causes conspicuous increase of the blood cholesterol, has no effect on the size of transplants.

FROM AUTHORS' SUMMARIES.

THE CULTURE OF WHOLE ORGANS. H. OKKELS, J. Exper. Med. **66**:297 and 305, 1937.

During the first and second day of cultivation of the whole thyroid gland in the Lindbergh apparatus, perfused by a medium containing 20 or 40 per cent homologous serum diluted with dextrose-Tyrode, the thyroid epithelium remains histologically normal and even the most delicate cytologic details retain their normal aspect. Cultivation over a period up to six days need not cause any serious histologic alteration of the gland, but cytologic examination reveals increasing damage of the mitochondria and of the Golgi apparatus. These changes seem not to interfere with the life of the cells, however; an epithelial culture from the perfused organs yields positive results. The process of cultivation is liable to cause a slight morphologic reaction in the thyroid epithelium, which resembles a feeble stimulation of the tissue. Addition of the thyroid-stimulating factor from the anterior lobe of the pituitary to the perfusion fluid causes a stimulation of the cultivated thyroid epithelium which is comparable to, but not so marked as, the corresponding reaction in the living animals.

Perfusion of isolated thyroid glands from rabbits, made refractory to the action of the thyroid-stimulating factor from the hypophysis by repeated injections for two months, has shown that in this condition the thyroid epithelium responds to pituitary stimulation. The serum from refractory rabbits does not neutralize completely the thyroid-stimulating effect of anterior pituitary extract on an isolated normal thyroid gland. It is believed that tissues other than the thyroid play a role in bringing about the phenomena which have been described as refractoriness against a prolonged action of the thyroid-stimulating pituitary factor.

FROM AUTHOR'S SUMMARIES.

INVESTIGATIONS ON THE BLOOD-BRAIN BARRIER. U. FRIEDEMANN, J. Immunol. **32**:97, 1937.

Intravenously injected basic dyes, with the exception of safranin, stained brain tissue readily in doses varying from 1 to 7 mg. Of acid dyes, some did not stain the brain at all, while others did, but only in larger doses. The affinity of the dye for brain tissue and the permeability of the brain capillaries, which is dependent on the electrical charge of the stain, are determining factors. Staphylococcus toxin, which has a negative charge at the p_H of the blood and the isoelectric point in the neighborhood of p_H 6.2, is unable to pass the blood-brain barrier. It occupies an intermediate position between cobra venom and lamb dysentery toxin, which pass the cerebral capillaries, and botulinus and tetanus toxins. The electrical charge alone proved insufficient to explain the differences in the behavior of the toxins. The differences of the ζ potentials of toxins are, according to Friedemann, better suited to explain the differences in the permeative capacity of the toxins. The ζ potential is the potential difference between the electrical double layer or the surface of the particles and the surrounding fluid. It depends on the chemical nature of the colloid and on the p_H of the fluid in which the colloid is dispersed.

I. DAVIDSOHN.

EXPERIMENTAL DUODENAL ULCER. I. F. VOLINI, H. L. WIDENHORN and B. FINLAYSON, Surg., Gynec. & Obst. **65**:159, 1937.

Utilizing the Exalto method and the Mann-Williamson procedures, the authors produced intestinal ulcers in all dogs subjected to one or the other of these operations. Heretofore the isolated segment of duodenum appears to have been overlooked by all investigators. Volini and associates report the occurrence of typical acute, subacute and chronic ulcers, varying in size and number, in the mucosa of the excluded duodenum. So far as they have been able to find, the formation of duodenal ulcers in the experimental animal has not been recorded in the literature. The duodenum of the normal dog contains punched-out depressions, grossly resembling ulcers but which microscopically prove to be normal intact mucosa. While a satisfactory explanation for these experimental ulcers is not at hand, trauma can be dismissed as the causative factor of those occurring in the duodenum.

WARREN C. HUNTER.

MORBID EFFECTS OF PITRESSIN (BETAHYPOPHAMINE) ON THE ORGANS AND VESSELS OF RATS. F. B. BYROM, J. Path. & Bact. **45**:1, 1937.

Large doses of pitressin injected repeatedly into the rat cause gross lesions in the kidneys, liver and other organs and in arteries. The renal lesions are certainly, the others probably, the result of focally intense arterial spasm, which causes (1) direct infarction or ischemic degenerative changes short of infarction of specialized parenchyma, (2) increased permeability of the vascular tree beyond the site of constriction and (3) necrosis of the artery itself when the spasm is maintained. These lesions are compared and contrasted with the changes observed in eclamptic toxemia, in certain etiologically related diseases and in "allergic" or "necrotizing" arteritis; the possible role of spasm in these diseases is discussed. The morbid effects of pitressin include the following eclampsia-like features: hypertension, delayed excretion of water, albuminuria, coma, catarrh and degeneration of the renal convoluted tubules, focal hepatic necroses (not histologically typical) and in pregnant rats death and maceration of the fetuses. The parallel is interpreted as supporting the view that the lesions of eclampsia and preeclampsia are expressions of vascular spasm, but reasons are given for rejecting the hypothesis that such spasm is caused by simple oversecretion of pitressin.

FROM AUTHOR'S SUMMARY.

RABIES VIRUS AND NEOPLASTIC CELLS. C. LEVADITI, R. SCHOEN and L. REINIÉ, Ann. Inst. Pasteur **58**:353, 1937.

The affinity of the street virus of rabies for the neurons and for the corneal epithelium is not restricted to these tissue systems. Cancerous cells of Pearce's tumor react in the same way. To the extent that these cells multiply in the anterior chamber of the eye of the rabbit the virus of rabies completes there its perfect cycle of evolution, including the development of the intracytoplasmic Negri bodies. From this point of view, the fixed virus of rabies behaves differently from the street virus. It is probable that the neoplastic elements lend themselves to the genesis of Negri bodies; that is, being in the embryonic state, they possess potentially certain fundamental properties of highly differentiated cells, such as those of the neurons and the corneal epithelium. FROM AUTHORS' SUMMARY.

INJECTIONS OF ACETONE EXTRACT OF THE TUBERCLE BACILLUS. L. P. H. J. DE VINK and J. H. TEN THIJE, Ann. Inst. Pasteur **58**:474, 1937.

Acetone extract of tubercle bacilli when injected into nontuberculous guinea pigs resulted in reactions in the reticuloendothelial system of the spleen and in the lungs. In the latter there appeared a thickening of the interalveolar spaces with proliferation of cells of the mesenchyme. In the lung and spleen a special type of cells was noted, with red protoplasm and a large nucleus, poor in chromatin

and round, oval or elongated. Several times giant cells were seen, differing from those of Langhans, the nuclei of which, usually 10 per cell, were scattered in the protoplasm, which was red in the section surrounding the nuclei.

RETICULOCYTES OF THE RABBIT: II. P. NICOLLE, Arch. Inst. Pasteur de Tunis **26**:326, 1937.

This, the second report of exhaustive studies, discusses reticulocyte counts made on the blood of rabbits during all phases of pregnancy. Estrogen produced reticulocytopenia; an extract of the anterior lobe of the hypophysis produced an increase in the number of these cells, and the urine of pregnant women produced vague periods of reticulocytopenia with an absence of typical reticulocytosis.

HETEROTOPIC PROLIFERATION OF THE GLANDS IN THE FUNDUS OF THE STOMACH STIMULATED BY INFLAMMATION. W. G. GARSCHIN, Frankfurt. Ztschr. f. Path. **49**:108, 1936.

In numerous rabbits, tar was injected subserosally into the fundus of the stomach and the response studied histologically at varying intervals. An area of necrosis was produced in the submucosa shortly after the injection. This was soon followed by the formation of an ulcer and inflammatory changes in the surrounding tissue. The neighboring fundic glands proliferated in two distinct ways, depending on their situation. Well differentiated glands were piled up outside of the inflammatory zone, whereas moderately undifferentiated glandular acini were formed within these zones. Many of the latter glands were seen in the submucosa. The author interprets this heterotopic hyperplasia as a response to an inflammatory stimulus and in no way neoplastic. He urges the use of criteria other than those of relatively slight invasion and lack of differentiation in the diagnosis of early neoplastic changes.

L. OHRINGER.

DIFFERENCES IN THE REGENERATING AND PROLIFERATING PROCESS OF THE RESPIRATORY TRACT. W. G. GARSCHIN, Frankfurt. Ztschr. f. Path. **49**:121, 1936.

Garschin studied regeneration of the epithelium of the respiratory tract after operative removal of the mucosa of the trachea in rabbits and inflammatory proliferation after injection of tar into the lungs. He points out that the regenerative and proliferative processes of the ciliated columnar epithelium in the respiratory tract are identical. In the beginning of the regenerative process there is substitution of the ciliated columnar epithelium by undifferentiated epithelium. The same occurs as the result of inflammatory proliferation. Later the undifferentiated cells close to the basal membrane give rise to ciliated columnar epithelium. In the next stages the undifferentiated cells of the upper layers also become columnar, so that the epithelium appears to be transitional. The squamous epithelium that appears in the regenerative and inflammatory proliferative stages of the stratified ciliated columnar epithelium is only an intermediate stage of the repair and must be differentiated from squamous cell metaplasia in the respiratory tract.

L. OHRINGER.

RELATION OF PREGNANCY CELLS OF THE HYPOPHYSIS TO GROWTH. J. ERDHEIM, Frankfurt. Ztschr. f. Path. **49**:452, 1936.

Pregnancy is accompanied by hypertrophy and hyperplasia of the chief cells of the anterior lobe of the hypophysis. These cells resemble morphologically the eosinophilic cells but are to be differentiated from them. In spite of postpartum involution of the chief cells (pregnancy cells), these remain conspicuous until the menopause. The significance of the pregnancy cells has been further obscured since the demonstration that they are not involved in the increased production of the gonadotropic factor during pregnancy. This hormone arises from the chorionic

epithelium of the placenta and from similar cells in instances of chorionepithelioma. The hypophysial changes characteristic of pregnancy have also been found in association with chorionepithelioma of the testes and in a 6 year old girl with chorionepithelioma of the ovary. Berblinger was able to produce similar changes in the hypophyses of rabbits by the use of placental extracts.

Two 6 year old girls with chorionepithelioma of the ovary showed precocious puberty and markedly increased skeletal growth. It seems that there is a causal relationship between the hypophysial changes (the presence of pregnancy cells) and growth and that the pregnancy cells secrete a growth factor.

Schmorl in 1933 reported the case of a 36 year old woman who died two years post partum, showing puerperal osteomalacia. There were marked proliferation of cartilage and renewed endochondrial ossification. Schmorl attributed these changes to hypersensitivity of the diseased cartilage to mechanical irritations. Erdheim, however, feels that such changes are caused by pituitary disorders. Three possibilities exist: There may have been present in the hypophysis an eosinophil adenoma, a pregnancy cell adenoma or hypertrophy of the chief cells as seen in chorionepithelioma. The last he feels is the most likely lesion. The hypophysis, it seems, was probably not examined, but Erdheim refers to the case in order to call attention to a possible relationship between skeletal and pituitary change.

A. STRAUSS.

Pathologic Chemistry and Physics

THE CALCIUM CONTENT OF THE KIDNEY AS RELATED TO PARATHYROID FUNCTION. W. DONOHUE, C. SPINGARN and A. M. PAPPENHEIMER, J. Exper. Med. **66**:697, 1937.

Partial nephrectomy in the rat leads to an increase in the calcium content of the residual renal tissue. This increase is correlated with enlargement of the parathyroid glands, the degree of enlargement depending on the severity of the renal lesion. Early removal of the parathyroid glands almost completely prevents this increase in renal calcium.

FROM AUTHORS' SUMMARY.

PROTHROMBIN DEFICIENCY AND THE BLEEDING TENDENCY IN LIVER INJURY (CHLOROFORM INTOXICATION). H. P. SMITH, E. D. WARNER and K. M. BRINKHOUS, J. Exper. Med. **66**:801, 1937.

The tendency to bleed in acute chloroform intoxication is due to deficiency in both plasma fibrinogen and plasma prothrombin. If the disorder is mild, no bleeding occurs. However, the prothrombin falls to rather low levels, although the fibrinogen falls only moderately. The tendency to bleed may also be produced by giving small repeated doses of chloroform. In such experiments, the hemorrhagic tendency is due to a deficiency in prothrombin alone. The fibrinogen level is unaffected. The relation of the hepatic injury to the plasma prothrombin level indicates that the liver is concerned in the manufacture of prothrombin. The formation of prothrombin appears to be more easily interfered with than the formation of fibrinogen.

FROM AUTHORS' SUMMARY.

FIELD STUDIES OF SELENIUM POISONING. M. I. SMITH and B. B. WESTFALL, Pub. Health Rep. **52**:1375, 1937.

During September 1936 a study was made of a selected group of 50 rural families in four counties of South Dakota and Nebraska, which are a highly seleniferous area. One hundred urine specimens from as many subjects were analyzed for selenium. The selenium concentrations for the whole group varied from 20 to 198 micrograms per hundred cubic centimeters. There was, as a rule, little variation in the urinary concentration of selenium for the several members of the same family or for the same person at different times, indicating that the

excretion level of selenium in man is a fairly reliable index of the availability of selenium and of the hazard to which he is exposed.

Outside of a high incidence of symptoms pointing to gastric or intestinal dysfunction and a few instances of apparent hepatic dysfunction, both probably the result of the continual ingestion of selenium, no other evidence of ill health was seen that could be ascribed to selenium with certainty.

Analysis of many samples of locally produced foodstuffs which enter directly or indirectly into the human dietary indicates that meat, eggs, milk and vegetables, when these are available, constitute the most important and most constant sources of selenium to which man is exposed in these regions in which selenium poisoning is endemic.

From Authors' Summary.

The Source of the Fat in Experimentally Produced Fatty Degeneration of the Heart. J. H. Dible and W. W. Gerrard, J. Path. & Bact. **46**:77, 1938.

From the study of the myocardium of the guinea pig poisoned with phosphorus, using as a control an animal in a similar state of nutrition, the authors conclude that the fatty change which may be produced is of the nature of an infiltration and that there is no reason to assume an unmasking of the essential fat of the cell.

From Authors' Conclusion.

Microbiology and Parasitology

Sites of Election for Negri Bodies in Rabies. E. S. Horgan and R. M. McKinnon, J. Hyg. **37**:340, 1937.

Comparative examinations of the hippocampus and midbrain in 37 brains from animals suspected of being rabid has shown the marked superiority of the hippocampus as a site of election for Negri bodies.

From Authors' Summary.

Experimental Intracerebral Inoculation for Identification of Different Types of Tubercle Bacilli. I. Bertrand, J. Bablet and F. Bloch, Ann. Inst. Pasteur **59**:231, 1937.

Solitary cerebral tubercles as found in man could not be reproduced by intracerebral inoculation of rabbits with tubercle bacilli. The local reaction and the course of infection depended on the type of bacilli and not on the virulence of the strain.

The type of tubercle bacilli could be rapidly and accurately determined by intracerebral inoculation of rabbits.

Human strains did not produce cachexia or paralysis within one month. Bovine and avian strains were fatal in less than a month. Avian bacilli caused an acute toxicosis infection within from twelve to twenty-five days, which was characterized by diffuse lesions. Bovine cultures produced large nodular, caseous lesions of the meninges and cerebral parenchyma.

Intracerebral inoculation of the rabbits with BCG was followed by the appearance of transitory foci of benign inflammation, which was not intensified by subsequent short wave treatment or by injection of virulent bovine bacilli.

From Authors' Summary.

Production of Vesicular Hepatitis in the Rabbit by Bacillus Perfringens. E. Dalous, J. Fabre, H. Pons and M. Cahuzac, Ann. Inst. Pasteur **59**:329, 1937.

Vesicular hepatitis as it occurs in man could not be reproduced in rabbits by intramuscular or intravenous injection of a culture of Bacillus perfringens (Clostridium welchii) or by injection of the culture into the genitalia or directly into

the liver. It could be produced by injecting into the mesenteric vein exudate from guinea pigs infected with cultures isolated from the soil.

Vesicular hepatitis caused by B. perfringens is the initial lesion of a rapidly fatal metastatic gas gangrene of the viscera due to infection of the liver by way of the portal system. It differs from other bacterial infections of the liver in which invasion is by the arterial route (described by Fiessinger as hepatitis of elimination).

J. B. GUNNISON.

CLINICALLY INAPPARENT EXPERIMENTAL SYPHILIS OF THE MOUSE. G. STROESCO and A. VAISMAN, Ann. Inst. Pasteur **59**:403, 1937.

Mice inoculated subcutaneously with chancre tissue from rabbits infected with Spirochaeta pallida were killed at intervals, and microscopic examinations were made by Stroesco's silver impregnation method.

The spirochetes invaded the cutaneous tissue and found optimal conditions for proliferation in the malpighian layer. They spread by way of the skin and peripheral nerves but did not invade the blood vessels and were not disseminated by the blood stream. They did not penetrate the striated muscle, transversing this layer only by way of the nerves.

After two months, spirochetes were demonstrated in the cutaneous layers of all parts of the body, all mucous membranes, the genitourinary organs (except the testicle), the salivary and mammary glands, the submucosa of the entire digestive tract, the periosteum of all bones, the nasal and pharyngeal cartilage, the peripheral nerves, the spinal and cerebral ganglions and the meninges. They did not penetrate into the parenchyma of the testicle, spleen, liver, lungs or brain. Multiplication and penetration did not occur in the central nervous system, although it was marked in the peripheral nerves.

No clinically or histologically demonstrable reaction accompanied this widespread penetration. The epidermal cells remained intact, and there were no manifestations of response to a foreign body or of toxic action. The spirochetes retained their virulence, as was demonstrated by inoculation of these invaders into the scrotal tissues of rabbits.

Localization did not follow embryologic laws; i. e., tissues and organs derived from the same germ layers were not necessarily invaded.

J. B. GUNNISON.

Immunology

PATHOGENESIS OF CORTICAL NECROSIS OF THE KIDNEYS IN RABBITS DUE TO STAPHYLOCOCCUS TOXIN. J. H. GLYNN, Am. J. Path. **13**:593, 1937.

Attention is called to two errors in previous studies on the effect of staphylococcus toxin on the rabbit's kidneys: (*a*) the failure to select animals on the basis of preliminary titrations of their serums, many of which possess considerable quantities of "natural" antitoxin; (*b*) the failure to measure toxin in terms of some unit of activity. Evidence based on mitochondrial changes in tubular epithelium is presented favoring the view that staphylococcus toxin exerts a direct effect on the kidney cells separate from the changes secondary to hemorrhage. Previous reports of vascular damage are confirmed, and the mechanism of hemorrhage is described. Cortical necrosis is eventually the result of two factors: (*a*) direct injury to cells by toxin and (*b*) anemia resulting from hemorrhage.

FROM AUTHOR'S SUMMARY.

VIABLE PNEUMOCOCCI AND PNEUMOCOCCIC SPECIFIC SOLUBLE SUBSTANCE IN THE LUNGS IN LOBAR PNEUMONIA. R. N. NYE and A. H. HARRIS, Am. J. Path. **13**:749, 1937.

Quantitative bacteriologic methods have shown that viable pneumococci are extremely scarce in the consolidated lobes of lungs from persons who have died of

lobar pneumonia. A quantitative serologic method has demonstrated that homologous soluble specific substance is present in such lobes in high concentration and that, as a rule, the lobes showing moderately old consolidations contain the largest amounts. Histologic examination of sections of the same lobes has shown that the majority of the pneumococci are phagocytosed by polymorphonuclear leukocytes.

FROM AUTHORS' SUMMARY.

IMMUNOLOGIC STUDIES ON PEPSIN AND PEPSINOGEN. C. V. SEASTONE and R. M. HERRIOTT, Am. J. Physiol. **20**:797, 1937.

Alkali (p_H 7.6)-denatured pepsins from swine, cattle and guinea pigs precipitate in swine pepsin antiserum. Similarly treated pepsins from the rabbit, chicken and shark do not. Pepsin antiserums react with both pepsin and pepsinogen but do not react with the serum proteins from the homologous species. Pepsinogen antiserums react with pepsinogen but not with twice crystallized pepsin or with the serum proteins from the homologous species. Positive reactions between activated pepsinogen and pepsinogen antiserum have been observed. It was possible to remove the reacting material from either the pepsinogen or the activated pepsinogen mixture. Antiserum made with serum proteins does not react with the homologous pepsin or pepsinogen.

FROM AUTHORS' SUMMARY.

TUBERCULIN ALLERGY PRODUCED BY PARENTERAL BCG VACCINATION. C. KERESZTURI, H. A. ROSENBERG and W. H. PARK, Am. Rev. Tuberc. **36**:90, 1937.

This is a report of the development of allergy to old tuberculin (0.1 and 0.2 mg.) in 292 intracutaneously and 41 subcutaneously BCG vaccinated children. Relatively more of the intracutaneously vaccinated group (80 per cent) presented a positive Mantoux reaction than of the subcutaneously vaccinated group (62 per cent), and sooner. The highest incidence of allergy in both groups occurred at the end of the sixth month. At the end of the first year the percentage giving a positive Mantoux reaction was 75 in the intracutaneously vaccinated group and 50 in the subcutaneously vaccinated group; at the end of the second year, it was 45 and 36, respectively, and at end of the third year, 42 and 30. In the intracutaneously vaccinated group a 0.15 mg. dose produced a greater incidence of allergy than any smaller dose. Larger doses showed no advantage. With the small doses used for subcutaneous vaccination, no effect on the incidence of allergy was noted; in 2 children with 0.05 mg. doses a positive Mantoux reaction developed, but such doses have the undesirable feature of producing local cold abscesses. The incidence of allergy was not influenced by the age of the patient at the time of vaccination. A direct relationship exists between the incidence of allergy and the severity of the local reaction and also between the incidence of allergy and the severity of the reaction of the regional lymph nodes. The age of the vaccine (from one to ten days) does not influence the incidence of the positive Mantoux reaction. A relatively larger number of children exposed to open tuberculosis before or after vaccination showed a positive Mantoux reaction than of those not exposed. There were some children who could not be made allergic to old tuberculin despite several vaccinations with BCG.

H. J. CORPER.

EFFECT OF OXYGEN INJECTED SUBCUTANEOUSLY ON ANTIBODY FORMATION. G. P. YOUMANS and T. SIMPSON, Arch. Int. Med. **60**:574, 1937.

A study was made of the effect of subcutaneous administration of oxygen on the production of agglutinin and opsonin in rabbits and dogs into which heat-killed type I and type II pneumococci had been injected and on the complement titers of rabbits. In this work 10 rabbits and 6 dogs received injections of heat-killed pneumococci of type I and type II. Half were given daily subcutaneous injections of 30 cc. of oxygen per kilogram of body weight, and the remainder

were used as controls. The opsonin and the agglutinin content of the serum were determined for all animals at frequent intervals over a period of three weeks after the last injection of antigen. Complement was titrated at the same time for all rabbit serums. The subcutaneous administration of oxygen had no effect on the opsonin content of the serum of dogs or rabbits for either the type I or the type II pneumococci. The subcutaneous administration of oxygen had no effect on the agglutinin content of dog serum for the type I and the type II pneumococci or on that of rabbit serum for the type II pneumococci. An apparent effect of the subcutaneous injection of oxygen on the agglutinin content of rabbit serum for the type I pneumococci was found statistically to be without significance.

FROM AUTHORS' SUMMARY.

IMMUNOLOGIC STUDIES OF SICKLE CELL ANEMIA. W. W. CARDOZO, Arch. Int. Med. **60**:623, 1937.

The term "sicklemia," as suggested by Cooley, is preferable to "sickle cell anemia" because the majority of persons whose red cells showed sickling were not anemic. The distribution of blood group factors A, B, AB and O and of blood types M, N and MN among 119 Negroes with sicklemia was similar to the distribution of these among 1,143 Negroes with normal blood and among 307 non-Negroes. The incidence of sicklemia was 9.42 per cent among the Negroes and 0.32 per cent among the non-Negroes (1 Mexican among 307 persons). An attempt to detect a specific agglutinogen in erythrocytes of the sickle form failed. There was no evidence of abnormality in the isoagglutinogens of the red cells or in the isoagglutinins of the serums. The property of sickling was found to be independent of the serum, and it remained with the cell as long as the latter was intact.

I. DAVIDSOHN.

IMMUNOLOGIC RELATIONSHIP BETWEEN THE SWINE AND HUMAN INFLUENZA VIRUSES IN SWINE. R. E. SHOPE, J. Exper. Med. **66**:151, 1937.

Swine that have recovered from either swine influenza or infection with swine influenza virus alone are usually not only immune but refractory to human influenza. Swine that have recovered from infection with a mixture of human influenza virus and Haemophilus influenzae suis are usually immune to swine influenza, while those that have recovered from infection with human influenza virus alone are usually not immune to swine influenza. The possible mechanisms involved in the cross immunity between the influenza viruses are discussed.

FROM AUTHOR'S SUMMARY.

EFFECT OF HAEMOPHILUS INFLUENZAE SUIS VACCINES ON SWINE INFLUENZA. R. E. SHOPE, J. Exper. Med. **66**:169, 1937.

Either living or heat-killed Haemophilus influenzae suis vaccines on being given intramuscularly to swine elicit an immune response capable of modifying the course of later swine influenza. The protection afforded is partial and in no way comparable to the complete immunity afforded by swine influenza virus vaccines.

FROM AUTHOR'S SUMMARY.

SPECIFICITY OF AZOPROTEIN ANTIGENS CONTAINING GLUCURONIC AND GALACTURONIC ACIDS. W. F. GOEBEL and R. D. HOTCHKISS, J. Exper. Med. **66**:191, 1937.

Azoprotein antigens containing glycuronic and galacturonic acids give rise in rabbits to specific antibodies. The immune serums show no serologic crossing with antigens containing dextrose or galactose. The galacturonic acid antigen reacts in antipneumococcus horse serum type I in high dilutions. Azoprotein

antigens containing galacturonic acid, benzene sulfonic and carboxylic acids precipitate in antipneumococcus horse serums of various types but not in normal horse serum. The mechanism underlying these cross reactions is discussed.

FROM AUTHORS' SUMMARY.

A NATURAL HEMOLYSIN FROM THE RAT PRODUCING NUCLEAR LYSIS OF CHICKEN ERYTHROCYTES. D. WEINMAN, J. Immunol. **32**:1, 1937.

This is apparently the first observation of a thermolabile hemolysin in rat serum for the erythrocytes of chickens. The lysis included the nucleis. No difference in hemolytic properties was noted between the serum of rats infected with Bartonella muris and that of splenectomized rats. A detailed study is reported of changes in the cytoplasm and nuclei of chicken red cells mixed with hemolyzing rat serum.

I. DAVIDSOHN.

ORIGIN OF NATURAL IMMUNE BODIES FOR PNEUMOCOCCI IN INFANTS. W. D. SUTLIFF and J. A. V. DAVIES, J. Immunol. **32**:43, 1937.

Nasopharyngeal cultures from 25 infants from 5 to 21 months of age were negative for pneumococci of types I, II and III. Tests for pneumococcidal action for the aforenamed three types of pneumococci in whole blood from 9 infants were negative at first but after three months such tests showed that the blood of 3 infants was pneumococcidal for type II and the blood of 1 for type III. At the same time bactericidal properties were detected in the blood of most of the examined infants for types IV, VI and XVII. Most of the infants had on various occasions positive throat cultures. In 59 per cent of these the type could not be determined but in 41 per cent the types were IV, VI, XV, XVII or XIX. There was no specific relationship between the strain recovered from the nasopharynx and the pneumococcidal action of the blood.

I. DAVIDSOHN.

REVERSE ANAPHYLAXIS IN RATS WITH SPECIAL ATTENTION TO RENAL DAMAGE. J. E. SMADEL and H. F. SWIFT, J. Immunol. **32**:75, 1937.

The serum of rabbits given injections of suspensions of rat kidney produced renal lesions in rats. The animals could not be desensitized to the so-called nephrotoxin. In contrast with this, small doses of serum of rabbits inoculated with rat serum produced no lesions in rats and desensitized them against doses of the antiserum which were fatal to nonprepared animals. The lesions in the rats given injections of large amounts of antiserum were vascular in origin and hemorrhagic and exudative in character. If death was instantaneous, the lesions were mainly in the lungs; if death was delayed, other viscera showed involvement.

I. DAVIDSOHN.

MUTUAL MULTIVALENCE OF TOXIN AND ANTITOXIN. H. EAGLE, J. Immunol. **32**:119, 1937.

In the presence of an excess of toxin, diphtheria antitoxin binds more of the toxin than would be expected by its neutralizing ability, and the resulting compound is toxic. In the presence of an excess of antitoxin, the toxin combines similarly with more than the neutralizing quantity of antitoxin, and the resulting compound is antitoxic. The precisely neutral compound is formed only in the presence of certain definite quantitative proportions of the two reagents. The observations are in better agreement with the hypothesis of Bordet on the toxin-antitoxin reaction than with the hypothesis of Ehrlich or Arrhenius and Madsen. Both reagents seem multipotent with respect to each other. I. DAVIDSOHN.

RELATION OF TEMPERATURE TO ANTIBODY FORMATION IN COLD-BLOODED ANIMALS. F. W. ALLEN and E. C. MCDANIEL, J. Immunol. **32**:143, 1937.

Normal frog serum contains no lysins for human blood. From the fact (*a*) that 56 frogs kept at room temperature responded to inoculations of human blood with the production of hemolysins up to a titer of 1:3, (*b*) that 79 frogs kept at refrigerator temperature and treated similarly did not produce antibodies and (*c*) that untreated controls at room and refrigerator temperatures did not produce antibodies, Allen and McDaniel conclude that the elevation of temperature was the only factor responsible for the formation of antibodies. I. DAVIDSOHN.

A "NORMAL ANTIBODY" IN MONKEY SERUM. M. WEICHSEL and H. SALFELD, J. Immunol. **32**:171, 1937.

The serum of normal monkeys and of monkeys convalescing from experimental poliomyelitis contained a complement-fixing antibody that reacted with alcoholic solutions of commercial lecithin. The serum of the monkeys convalescing from poliomyelitis fixed complement in higher dilutions of the hapten than did serum of normal animals. Lecithin prepared from eggs failed to react with the serum. An acetone-soluble substance isolated from commercial lecithin which differed from lecithin in its chemical reactions fixed complement with serum of all monkeys.

I. DAVIDSOHN.

INFLUENCE OF NARCOTICS ON ANAPHYLACTIC SHOCK. L. FARMER, J. Immunol. **32**:195, 1937.

Ethyl carbamate protected 50 per cent of sensitized guinea pigs against anaphylactic shock. Morphine did not. The results confirm the findings of Besredka concerning the antianaphylactic action of certain narcotics. The explanation of Besredka that the effect is due to action on the central nervous system is held untenable by the finding that some of the narcotics act directly on the smooth muscles of the bronchi. The failure of morphine to protect is due to the fact that instead of relaxing the bronchial muscles it constricts them and thus acts in the same sense as the anaphylactic shock itself. I. DAVIDSOHN.

QUANTITATIVE CAPSULE SWELLING TESTS IN BLOOD SERUM OF PATIENTS WITH PNEUMONIA. J. G. M. BULLOWA and J. SHARFF, J. Infect. Dis. **61**:55, 1937.

Clapp's method of determining quantitatively the substance which causes capsule swelling in the pneumococcus is described. This antibody appears in the serum when patients recover from pneumonia. The results obtained by Clapp's method parallel the estimates of protective antibody made by the more costly and time-consuming mouse protection test.

A patient with fatal type II pneumococcic pneumonia had bacteremia simultaneously with the presence of the substance causing capsule swelling.

Because bacteremia may be present coincidently with the presence of this substance and of agglutinin in the blood of patients suffering from type II pneumococcic pneumonia, these antibodies are not satisfactory guides to adequacy of treatment.

The amount of the substance which causes capsule swelling in the blood of patients recovering from pneumonia, either spontaneously or after treatment with specific serum, may be determined, provided serum of known mouse protection titer and fresh cultures of pneumococci with good capsules are available.

FROM AUTHORS' SUMMARY.

EXPERIMENTAL INFECTION WITH CLOSTRIDIUM OEDEMATIS MALIGNI. D. W. HENDERSON, Brit. J. Exper. Path. **18**:224, 1937.

A heat-labile bacterial antigen prepared from vibrion septique has been shown to stimulate the production in rabbits of an antibody which controls experimental infection in mice. The experimental evidence points to an association and perhaps identity of this antigen with the flagellar substance. On this theory the antigen is the H antigen and the protective antibody the H antibody. The relative value of an antitoxic and an antibacterial serum has been examined, and the practical application of an antibacterial prophylaxis has been indicated.

FROM AUTHOR'S SUMMARY.

THE COMBINING PROPERTIES OF VACCINIA VIRUS WITH THE ANTIBODIES DEMONSTRABLE IN ANTIVACCINIAL SERUM. M. H. SALAMAN, Brit. J. Exper. Path. **18**:245, 1937.

The hypothesis is tentatively advanced that neutralization of the virus by antivaccinial serum depends on an antibody distinct from those responsible for agglutination and precipitation.

FROM AUTHOR'S SUMMARY.

CORYNEBACTERIUM PYOGENES, WITH SPECIAL REFERENCE TO TOXIN PRODUCTION. R. LOVELL, J. Path. & Bact. **45**:339, 1937.

This investigation and those made by other workers show that Corynebacterium pyogenes is a pleomorphic gram-positive rod which grows under aerobic and anaerobic conditions and in an atmosphere containing 10 per cent of carbon dioxide. It prefers a medium containing blood or serum, though this is not essential for growth; it liquefies gelatin, coagulated serum and egg; it clots and digests milk, produces beta hemolysis in blood agar plates and ferments several carbohydrates without the formation of gas. Serologic investigations have shown that five strains of C. pyogenes which have been cultured for some years contain a major and a minor antigen. Most strains are homogenous and possess only minor antigen. The studies showed no difference between strains isolated from different species of animals or from different disease processes. C. pyogenes produces a heat-labile hemolysin and a toxin lethal to rabbits and mice on intravenous inoculation. There is a correlation between hemolytic activity and toxicity, pointing to their identity. The toxin is relatively weak, is antigenic and may be converted to a nonhemolytic toxoid by a 40 per cent solution of formaldehyde.

FROM AUTHOR'S SUMMARY.

HEREDITARY TRANSMISSION OF M AND N. E. PONZI, Riv. ital. di ginec. **19**:462, 1936.

Ponzi determined the MN types of 100 pairs of parents and their children and did not encounter a single exception to the theory of heredity of Lansteiner and Levine.

A. S. WIENER.

SEROLOGIC INVESTIGATIONS OF VENEREAL LYMPHOGRANULOMA. H. HILDEBRAND, Zentralbl. f. Bakt. (Abt. 1) **137**:245, 1936.

Rabbits were immunized over long periods with the killed virus of venereal lymphogranuloma obtained from emulsions of the brains of infected mice. Specific complement-fixing antibodies were easily demonstrated in serum of these rabbits. On the other hand, complement fixation tests with serum from patients with venereal lymphogranuloma were for the most part negative. Neutralizing antibodies were not demonstrable in the serum of either infected patients or rabbits

immunized against the virus. These investigations indicate that the tendency of venereal lymphogranuloma to heal is slight and suggest, according to the author, that the mechanisms of immunity are depressed. PAUL R. CANNON.

ACTIVE IMMUNIZATION OF CATTLE AGAINST FOOT AND MOUTH DISEASE WITH FORMALDEHYDIZED VIRUS. O. WALDMANN, K. KÖHE and G. PYL, Zentralbl. f. Bakt. (Abt. 1) **138**:401, 1937.

The authors suspended the virus of foot and mouth disease in a phosphate buffer solution containing formaldehyde and tested its immunizing potentialities in about 2,000 guinea pigs and 300 young cattle. It was possible to immunize almost all the guinea pigs to the extent that only a primary lesion developed, with no evidence of generalization. About two thirds of the cattle also acquired definite immunity, which was demonstrable for at least four weeks and was not tested after a longer period. The authors do not believe that the formaldehyde completely inactivated the virus but suggest that it changed it so that when administered it was subinfectious, producing relatively solid immuntiy. PAUL R. CANNON.

IRREGULAR HEMAGGLUTINATION IN DISEASES WITH SPASTIC VASCULAR OCCLUSION. P. NEUDA, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:199, 1936.

Neuda observed and reported previously an autoagglutination reaction observed in the serums of patients with angina pectoris, intermittent claudication, Raynaud's disease and peptic ulcer, respectively. Incubation of the autoagglutinating serum for twenty-four hours destroyed the autoagglutinating property but brought forward the blood group factor. Inactivation preserved the autoagglutinating property and prevented the appearance of the blood group factor. The present report deals with the findings of irregular isoagglutination in 3 cases. The inactivated serum of a patient with Raynaud's disease and migraine, group $O\alpha\beta$, agglutinated A_1 cells feebly and A_2 cells strongly. The incubated serum agglutinated A_1 and A_2 cells equally well. The result indicated the following irregularity: inhibition of the agglutination of A_1 cells but not of the agglutination of A_2 cells by the inactivation. Inactivation had also some, though slight, inhibiting effect on the β isoagglutinin. Absorption of the incubated serum with A_1 cells did not remove the anti-A_2 agglutinins; similar absorption of inactivated serum removed the agglutinin for A_2 cells that reacts at 37 C. but not that which reacts at low temperatures. Absorption of inactivated serum with A_1 cells removed all agglutinins for A_2B cells, while absorption of incubated serum did not affect the same agglutinins. The serum of another $O\alpha\beta$ patient with migraine behaved similarly though not identically. The serum of a third patient, group A_2B, agglutinated A_1 erythrocytes.

I. DAVIDSOHN.

COMPLEMENT IN TOXIC ACTION OF FORSSMAN ANTISERUM ON CHICK EMBRYOS. O. G. BIER and E. SEILER, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**: 211, 1936.

Serum containing Forssman antibodies when added to chick embryos about 5 days old damaged and eventually killed them. The damage was due to the reaction between the Forssman antigen in the embryos and the introduced antibodies. In this reaction complement is a necessary factor and even the same fractions of complement (third and fourth) as those that are needed for the hemolysis of sheep red cells by specific antiserum. Chicken complement can reactivate the Forssman antiserum if sufficiently large doses are used.

I. DAVIDSOHN.

So-Called Fractions of Complement in the Wassermann Reaction. O. G. Bier, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:218, 1936.

Guinea pig complement in which the third or fourth fractions had been inactivated with yeast or with ammonia when added to syphilitic serum changed the Wassermann reaction from positive to negative. A similar effect followed the addition of guinea pig serum that had been heated for five minutes at 56 C. If, however, the heat-inactivated complement was added after an interval of one hour, the positive reaction remained unchanged. Complement that was inactivated for five minutes at 56 C. was partially reactivated after a stay in the ice box. The ability of guinea pig complement inactivated by heat or by yeast or ammonia to change complement fixation from a positive to negative was utilized by Bier to develop a quantitative procedure for the complement fixation test for syphilis. He admits that there is a possible source of error in the anticomplementary action of ammonia.

I. Davidsohn.

The Antigenic Functions of Hormonal Preparations. V. Kindermann and F. Eichbaum, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:230 and 498, 1936.

Rabbits were given repeated injections of the urine of pregnant women. This treatment did not interfere with the formation of corpora lutea in the female animals. The animals showed as a result only serologically demonstrable species-specific antibodies (complement-fixing and precipitating antibodies) against human serum. In a previous report, Kindermann and Eichbaum showed that after a sufficiently large number of injections hormone-inactivating substances developed and that eventually the serologic antibodies disappeared, while the antihormones persisted. The hormone-inactivating substances were also species specific.

Rabbits were given repeated injections of an extract of the anterior lobe of the pituitary of the cow or the pig, containing the thyrotropic hormone. There developed complement-fixing antibodies and precipitins and at the same time hormone-neutralizing substances, all of which were very labile in vitro and disappeared quickly from the circulation in the animals. The antibodies were species specific as well as hormone specific, as was demonstrated by removal of the species-specific antibodies from the immune serums. The antihormonal action of the immune serums of rabbits and guinea pigs could be transferred passively to untreated animals. The antihormonal action was strictly species specific.

I. Davidsohn.

Differentiation of Human Red Corpuscles with Polyvalent Hetero-Immune Serum. F. Ottensooser, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:260, 1936.

The serum was produced by inoculating rabbits with mixtures of human blood of all groups and types. The serum of 2 of the 9 immunized rabbits showed a striking individual specificity when a mixture of serum from these two was absorbed with the red cells of one person: The absorbed serum reacted only feebly or after a longer interval with the cells of the same person but distinctly and after a shorter interval with the cells of other persons. The time that elapsed until agglutination was noticeable was used as an indicator of the degree of the removal of the agglutinins. Ottensooser considers the results as signifying that he has found an identifying (i. e., individual) reaction of man.

I. Davidsohn.

Immunity in Syphilis. J. Vásárhelyi, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:296, 1936.

Sixteen rabbits were successfully inoculated intratesticularly with suspensions of Spirochaeta pallida. After varying intervals they were treated with large amounts of neoarsphenamine. After ninety days attempts to reinfect the same

animals were not successful. Normal control animals similarly treated with neoarsphenamine became infected under the same conditions, showing that this treatment was not able to protect them. Transplantation of organs of the treated animals to healthy rabbits showed the presence of spirochetes in 4 of the treated animals. Vásárhelyi concludes that true spirochetocidal immunity develops in experimental syphilis of the rabbit.

I. DAVIDSOHN.

ALCOHOLIC PRECIPITATES OF AQUEOUS POLLEN EXTRACTS. P. MANTEUFEL and G. KÜHNE, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:362, 1936.

Guinea pigs were sensitized with alcoholic precipitates of aqueous extracts of pollens of Alopecurus pratensis. Reinjections after not less than two months and preferably after from six to eight months produced typical anaphylactic shock with occasional deaths, and typical changes were found at autopsy. The alcoholic precipitate was not suitable for producing the anaphylactic phenomena by the inhalation method of Manteufel and Preuner and was not able to produce the Arthus phenomenon or the Prausnitz-Küstner reaction. The blood serum of the sensitized guinea pigs did not sensitize other guinea pigs. Untreated guinea pigs from 9 to 11 months old, born of mothers that were sensitized from four to five months before their birth, manifested typical anaphylactic shock when given intracardial injections of the homologous alcoholic precipitates, showing that hypersensitiveness of guinea pigs transmitted through the placenta may last at least two hundred and seventy days and not only seventy days, as had been assumed. Attempts at sensitizing guinea pigs with alcoholic precipitates of aqueous extracts of a mixture of sixteen different pollens resulted only in mild reactions.

I. DAVIDSOHN.

THE TUBERCULOUS NATURE OF RHEUMATIC FEVER. P. MANTEUFEL and G. DEMMER, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:373, 1936.

The Meinicke precipitation reaction and Mueller's immune ball reaction for tuberculosis, with addition of the extract of Witebsky, Klingenstein and Kuhn, were determined on bloods of 66 patients with various rheumatic diseases (acute rheumatic fever [active and resting], secondary chronic polyarthritis with a background of rheumatic fever, and primary chronic polyarthritis); 39.4 per cent of them and more than 50 per cent of patients with arthritis deformans reacted positively. Patients with acute rheumatic fever reacted negatively. The tests were properly controlled, and tested subjects were known to react positively only if they had active tuberculosis and to give false positive results only rarely. There were no clinical differences between the patients with rheumatic diseases whose tests were positive for tuberculosis and those whose tests were negative. Those reacting positively had no demonstrable tuberculous foci. Manteufel and Demmer conclude that there is no way of excluding the possibility of an etiologic relationship between tuberculosis and some rheumatic diseases.

I. DAVIDSOHN.

THE ORIGIN OF ISOAGGLUTININS IN MAN. O. THOMSEN, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:435, 1936.

After a review of the various hypotheses dealing with the subject, Thomsen argues in favor of the concept of the hereditary nature of the isoantibodies. The relation of the antibody genes to the genes of blood group factors is presented as a working hypothesis.

I. DAVIDSOHN.

BLOOD GROUPS IN RABBITS AND SPECIFICITY OF ISO-IMMUNE SERUM. P. V. MARCUSSEN, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:453, 1936.

Cross immunizations of 42 rabbits with the bloods of other rabbits led to the formation of isoimmune serums. Proper absorptions demonstrated the existence

of two main receptors in the blood of rabbits in addition to several less easily demonstrable antigenic structures. No rabbit was free from both receptors; 18 rabbits had one, 20 had the other, and 4 had both. A minority of the immune serums (8 of 41) were monovalent, 17 were polyvalent, and 6 were defective. These properties showed change in the course of immunization.

I. DAVIDSOHN.

A PSEUDOSYPHILITIC REACTION. I. JACOBSOHN, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:511, 1936.

Cultures of Bacillus subtilis on normal serum or on broth fixed complement with cholesterolized beef heart extract in a manner similar to that of syphilitic serums. Adsorption and dilution experiments with the cultures of B. subtilis and with syphilitic serums and the use of different fractions of the beef heart extract failed to demonstrate any differences in the behavior of the cultures of B. subtilis and syphilitic serums. Alcoholic extracts of B. subtilis retained the ability to fix complement with the beef heart extracts though they did not as regularly as the cultures of the organism. The reacting substance was found in the solution, as evidenced by the ability of the Berkefeld filtrate to act in a manner similar to that of the whole culture, while suspensions of the bacteria in saline solution failed to fix complement.

I. DAVIDSOHN.

SUBGROUPS OF GROUP B. M. MASAKI, M. KUWASHIMA, S. ISEKI and T. FUKAO, Hanzaigaku-Zasshi **9**:467, 1935.

These authors present experimental results which seem to show that there are differences among bloods of group B with regard to their ability to absorb anti-B agglutinins. On the basis of these quantitative differences a subdivision of the bloods of group B into two subgroups, B_1 and B_2, analogous to the subgroups of group A (A_1 and A_2) is proposed.

A. S. WIENER.

IDENTIFICATION OF TWO GROUPS OF HUMAN BLOOD CELLS WITH THE AID OF EEL SERUM. S. SUGISHITA, Juzenkai-Zasshi **40**:1938, 1935.

The author found that the serum of some eels reacts with erythrocytes of all four blood groups with equal intensity. The serum of others, however, reacts more strongly with O cells (titer, 1:8,000) than with A, B and AB blood cells. By the agglutinating activity of such serum for A, B and AB cells bloods can be arbitrarily divided into two types. Those which are agglutinated by higher dilutions of the serum (up to 1:4,000) are said to belong to type E, and those which are agglutinated only by undiluted or at least not highly diluted serum (less than 1:40) are said to belong to type e. By adjusting the dilution of the eel serum properly it may be possible to differentiate bloods of type E from those of type e directly by simple agglutination tests.

A. S. WIENER.

Tumors

GONADOTROPIC HORMONES AND CANCER: SOME EFFECTS OF THE INTERPLAY OF SUCH HORMONES ON THE INCIDENCE OF MAMMARY CANCER IN MICE. W. S. MURRAY, Am. J. Cancer **30**:517, 1937.

From these data, it seems that the introduction of the androgenic hormones into the blood stream of female mice interfered with their sexual cycle. The ovaries were stimulated to a precocious and prolonged production of follicles, which degenerated within the gland, with the result that no luteal tissue was formed. Since mammary tumors did not develop in these females, it seems reasonable to assume that the proliferation and change to malignant growth in the mammary

glands may have been due in some measure to the luteal fraction of the estrogenic hormone. On this hypothesis, the observed difference in cancer incidence between the breeding and the virgin females may be explained as due to the prolonged luteal phase of pregnancy. This explanation is also supported by the observation of Lacassagne, who found a correlation between the rapidity of normal estrous cycles and the incidence of cancer in mice.

From Author's Summary.

Effect of Gamma Radiation on Mitosis. S. Warren, Am. J. Roentgenol. **38**:899, 1937.

Radon does not differ essentially from roentgen radiation in its effect on mitosis. The premitotic phase is most sensitive to irradiation by radon as shown by the arrest of mitosis in Walker rat carcinoma 256. The drop in mitotic count occurs rapidly during the first five hours after the onset of irradiation, and the count turns toward normal, though usually remaining lowered, about seven hours later. Results reported here as obtained with tumor grafts are similar to those obtained with tissue culture material reported by others.

From Author's Conclusions.

Episacroiliac Lipoma. Emil Ries, Am. J. Surg. **34**:490, 1937.

A previously undescribed clinical type of lipoma occurring in a special location over the sacroiliac joint is established, to which the name "episacroiliac lipoma" may properly be applied. The connection of this type of tumor with backache, the surgical treatment for it and the results deserve wider attention.

From Author's Summary.

Primary Carcinoma of the Trachea with Cutaneous Carcinomatosis. E. L. Keeney, Bull. Johns Hopkins Hosp. **61**:411, 1937.

Cutaneous carcinomatosis may occur whenever a fascial lymphatic plexus is invaded by a neoplasm, and it is reasonable to believe that the parietal dissemination of the tracheal carcinoma which occurred in the patient reported on developed in such a way. The results suggest that if cutaneous metastases occur in a case of tracheobronchial obstruction, the primary growth is more likely to be located in the trachea than in the parenchyma of the lungs or in the bronchi.

Lung Tumors in Mice. H. B. Andervont, Pub. Health Rep. **52**:1584, 1937.

When 1,2,5,6-dibenzanthracene was injected subcutaneously into strain A mice, in different mediums, namely, dispersed in horse serum, dissolved in lard, suspended in glycerin, and dissolved in cholesterol, it was found that the substance dispersed in horse serum induced the most pulmonary growths.

Pulmonary tumors were induced in mice of strain A, strain C_3H and strain C57 black by inserting 1,2,5,6-dibenzanthracene directly into their lungs.

From Author's Summary.

The Serologic Inter-Relationships of the Viruses of Rabbit Myxomatosis and Fibroma. J. C. G. Ledingham, Brit. J. Exper. Path. **18**:436, 1937.

Rabbits infected with the virus of fibroma acquire agglutinins in their serums which act on both myxoma and fibroma. On the localizing action in vivo of such agglutinins probably depends the resistance to the superposition of an infection with myxoma exhibited by animals that have recovered from fibroma. Rabbits which recover, as they may do, though rarely, from an infection with myxoma naturally contracted and rabbits infected with the neuromyxoma variant of Hurst contain in their serums agglutinins for both myxoma and fibroma. The viruses

of fibroma and myxoma are probably not identical, serologically analyzed, the former being distinguished by its lack of an antigen fraction capable of stimulating the formation of myxoma-neutralizing antibodies.

FROM AUTHOR'S SUMMARY.

THE AGENT OF THE ROUS FOWL SARCOMA. E. M. FRAENKEL and C. A. MAWSON, Brit. J. Exper. Path. **18**:454, 1937.

Concentration of the active agent of the Rous fowl sarcoma is not invariably obtained in the deposit obtained by fractional centrifugation for two hour periods at 15,000 revolutions per minute. There is no relation between the tumor-producing activity of a preparation and the number of "elementary bodies" visible therein. A small variation in the quantity of Rous fowl sarcoma extract injected produces differences in the size of tumors and in the time of their appearance in the same bird, pointing to the chemical nature of the agent. No agent could be obtained by extraction with acetone and carbon tetrachloride; the extracts obtained by these procedures seemed to contain an inhibitor; the agent was obtained, however, from the residue remaining after extraction of the dried Rous fowl sarcoma tissue.

FROM AUTHORS' SUMMARY.

A FURTHER STUDY OF TERATOMA. R. A. WILLIS, J. Path. & Bact. **45**:49, 1937.

This paper describes the structure of ovarian teratoma as observed in 14 new cases. The structure was studied in serial sections. The results confirm and amplify those reached in a previous study. The nomenclature of teratoma is overburdened and the classification of the cases into various subgroups is artificial and often misleading. The name "dermoid cyst" as applied in a case of teratoma is particularly unsatisfactory and should be discarded. A "dermoid cyst" is merely benign teratoma in which one of the skin-lined cavities has become relatively large. "Multiple dermoid cyst" is almost always a misnomer, the separate skin-lined cysts being only parts of a single tumor. Between innocent and malignant teratoma no sharp line of distinction is possible; all gradations occur. The malignancy of teratoma is often polyvalent; i. e., it involves many or all of the component tissues and not merely a single one. There are reasons for believing that previously benign, quiescent teratoma may suddenly assume active growth and exhibit polyvalent malignancy. Occasionally ordinary squamous cell carcinoma arises in the skin of otherwise benign cystic teratoma. An instance of this is described in which the widespread distribution of the carcinoma in the skin of the cyst suggested that the cyst contents may have developed carcinogenic powers. When any future opportunity occurs of obtaining a growth of this kind in the fresh state, the cyst contents should be saved and tested for carcinogenic activity. Although correlation of tissues can many times be discerned in teratoma, the growth is fundamentally nonfetiform. It shows no signs of axiation, somatic architecture or regional organ formation. The mode of origin of teratoma is still obscure, but the solution of the problem will undoubtedly come with increasing knowledge of organizer activity and of the chemical processes of the embryo.

FROM AUTHOR'S SUMMARY.

THE INDUCTION OF TUMORS BY METHYLCHOLANTHRENE IN MICE. E. BOYLAND and F. L. WARREN, J. Path. & Bact. **45**:171, 1937.

The C. B. A. (agouti) strain of mice is less susceptible to the carcinogenic action of methylcholanthrene injected subcutem than the Simpson strain of aibino mice. The susceptibility appears to be the same in both sexes and in normal and castrated mice. There is some evidence to show that the relation of dose to response is that of a normal drug reaction, which suggests that carcinogenic action is not of an anaphylactoid nature.

FROM AUTHORS' SUMMARY.

INFLUENCE OF ESTROGEN ON THE GROWTH OF MALIGNANT CELLS. M. D. GILMOUR, J. Path. & Bact. **45**:179, 1937.

From the results obtained by applying estrogen to mice inoculated with transplantable tumors, it is concluded that this gonadotropic substance does not accelerate the growth of malignant cells, even of the mamma. It increases the susceptibility of the skin to the development of cancer in response to the carcinogenic agent 3,4-benzpyrene, and it hastens the appearance of tumors.

FROM AUTHOR'S SUMMARY.

ACTION OF COBRA VENOM ON EHRLICH'S SARCOMA OF MICE. A. BESSEMANS and L. ASAERT, Ann. Inst. Pasteur **58**:247, 1937.

Immersion of 0.1 cc. of tissue extract of Ehrlich's sarcoma in a solution of 0.02 mg. of a preparation of cobra venoum for twenty-four hours at 6 C. or at 17 C. rendered an otherwise fatal inoculum avirulent. Of 20 mice, 5 showed progressive necrosis and disappearance of the sarcoma when given a fatal subcutaneous dose followed immediately by 0.02 mg. of the cobra venom in 0.5 cc. of solution at the same site. Later injections failed to reduce the tumor.

ADVANCES IN EXPERIMENTAL CANCER RESEARCH. K. H. BAUER, Arch. f. klin. Chir. **189**:123, 1937.

This paper was presented before the sixty-first annual meeting of the German Society of Surgery on March 31, 1937. It is a comprehensive sixty-two page review of the problems of experimental cancer research. Its four chapters deal with (1) transplantable tumors, (2) inheritance of human and animal tumors, (3) carcinogenic substances and (4) the action of carcinogenic substances on tumors of man. Bauer reports his own experiences with 1,2-benzpyrene in the treatment of 22 carcinomatous persons. The chemical was applied in an ethereal solution to the surface of the growth or was injected. Seven patients remained tumor free for two years. In all cases the condition was cancer of the skin.

I. DAVIDSOHN.

CAUSES OF PRIMARY CARCINOMA OF THE LUNG. B. FISCHER-WASELS, Frankfurt. Ztschr. f. Path. **49**:145, 1936.

Since 1900, especially following the World War, there has been noted a universal increase of primary carcinoma of the lung, although the incidence of all malignant tumors has decreased. Fischer-Wasels comes to no definite conclusion regarding any one outstanding etiologic factor. He points out that in some cases primary carcinoma of the lung follows regenerative changes occurring in a tuberculous cavity or is present in lungs which revealed postinfluenzal bronchitis or bronchitis following measles. Pneumonoconiosis, irritations due to inhalation of nitrogen and those due to inhalation of poison gas may also be considered primary factors. Analysis of the causes of primary carcinoma of the lung in 52 per cent of the workers in the cobalt mines of Schneeberg who died from this cause led the author to conclude that the composition of the ore played no role. The radium emanation in the air within the cobalt mines seems to be the responsible agent.

A. STRAUSS.

TWO PRIMARY CARCINOMAS IN AN ANILINE WORKER. A. MÜLLER, Schweiz. med. Wchnschr. **17**:1031, 1936.

A squamous cell carcinoma of the urinary bladder was removed by resection from a 42 year old aniline worker who had been complaining of hematuria. No recurrence of the tumor was evident until five years later. Then hemorrhage developed from a tumor located in a double kidney on the left side. Death did not occur until eleven years after the operation on the bladder. Autopsy disclosed

in the left kidney a necrotic anaplastic carcinoma, which grew in the lumen of the whole length of the left ureter and which metastasized to the spleen, liver, lungs, hilar glands and bones and showed a histologic appearance different from that of the tumor of the bladder.

FREDERICK STENN.

THE SHOPE INFECTIOUS FIBROMA OF RABBITS. E. PASCHEN, Zentralbl. f. Bakt. (Abt. 1) **138**:1, 1936.

Paschen reports finding elementary bodies in cutaneous scrapings from a rabbit infected with the virus of infectious fibroma (Shope). These bodies were about 0.15 micron in diameter. They occurred both in an inflammatory strain of the virus and in a fibromatous strain. The inflammatory strain developed best in the superficial layers of the skin, whereas the fibromatous strain developed better in the deeper layers.

PAUL R. CANNON.

EFFECT OF GARLIC AND OF BACILLUS COLI ON TRANSPLANTED TUMORS. L. MAKOWSKY, Ztschr. f. Immunitätsforsch. u. exper. Therap. **89**:423, 1936.

In mice inoculated with Ehrlich's mouse cancer and fed garlic in addition to the regular diet, the tumors grew less rapidly than in the control animals and the garlic-fed animals lived considerably longer than the control animals, which died on the average after thirty-two days. The growth was from 33 to 40 per cent less rapid in the animals on the garlic diet, and in 6 animals the cancer implants did not take at all, while they took in all the 20 controls. The second part of the report deals with the influence of a change in the bacterial flora in the intestines, induced by feeding colon bacilli, on the growth of the same transplanted tumors. No effect was noted, but when garlic was subsequently fed to these animals an increase of resistance to the growing tumors was observed.

I. DAVIDSOHN.

OCCUPATIONAL OCCURRENCE OF CANCER OF THE LUNG. S. KURODA and K. KAWAHATA, Ztschr. f. Krebsforsch. **45**:36, 1936.

The writers have observed an excessive occurrence of pulmonary cancers in workers in gas plants, limited to those engaged at the generators. This work entails exposure to continued inhalation of hot tar-laden gas. An effort is being made to obtain similar results experimentally.

H. E. EGGERS.

THE INCREASE IN MORTALITY FROM CANCER. E. FÜRTH, Ztschr. f. Krebsforsch. **45**:47, 1936.

In a detailed analysis of German statistics on the mortality from cancer, particularly for the years 1926 and 1934, Fürth finds marked local discrepancies in reported death rates, changes of rate, age distribution and other items. The usual explanation of the apparent increase of mortality from cancer, that of the shift of age to higher levels, is entirely inadequate when the statistics are analyzed in sufficient detail. Nor, in Fürth's opinion, can any considerable significance be attached to such factors as changing dietary or greater diagnostic accuracy. In his opinion, the increase in the number of reported deaths represents an actual increase, the more striking in view of the increased efficiency in the treatment of superficially accessible cancer.

H. E. EGGERS.

Society Transactions

NEW YORK PATHOLOGICAL SOCIETY

ALFRED PLAUT, *President*

Feb. 24, 1938

ROBERT A. MOORE, *Secretary*

AN UNUSUAL CASE OF MYOCARDIAL CALCIFICATION WITH SUBSEQUENT INFECTION. MAURICE N. RICHTER.

The patient was a woman 43 years of age, whose illness extended over a period of one year, with mild cardiac symptoms, terminating with streptococcic septicemia.

The particular feature of interest was the heart. This was of normal size. A soft, friable vegetation was present on the aortic valve, but the cusps showed no evidence of previous disease.

In the endocardium and myocardium of the left auricle were numerous soft grayish nodules, those in the endocardium projecting into the lumen. These nodules contained yellowish or grayish white creamy material. Similar material, but less globular in form, was seen in the endocardium and myocardium of the left ventricle, and a small amount was also observed in the right auricle.

Microscopically, the masses were deposits of calcium. In the small masses the calcium was deposited in necrotic muscle fibers. Little cellular reaction was noted around the deposits. Some of the calcium deposits were infected, however, and contained pus with many gram-positive streptococci. A few focal inflammatory areas with polymorphonuclear cells and streptococci were present, without calcium deposits. The aortic vegetation contained numerous bacterial colonies, many polymorphonuclear and a few mononuclear cells.

The organism recovered from the lesions in the heart was identified as Streptococcus, group B, type II (Lancefield). This is an unusual organism to be found as a cause of fatal endocarditis. As it is hemolytic when grown anaerobically and nonhemolytic (with greenish colonies) when grown aerobically, its identity was uncertain until precipitin tests were made. The only other lesions of interest were chronic cystitis and bilateral hydronephrosis. No calcium deposits were observed other than in the heart.

One may consider as offering possible explanations for the calcium deposits (1) renal damage, (2) some previous toxic damage to the heart or (3) infection by the group B streptococcus obtained in this case. In any case, the occurrence of calcium within necrotic muscle fibers suggests that its deposition followed toxic necrosis of the myocardium rather than abscess formation or endocarditis and that infection of the calcified areas occurred secondarily.

DISCUSSION

ROBERT A. MOORE: I should like to ask Dr. Richter whether he regards this case as an instance of healing subacute bacterial endocarditis. Dr. Ferris recently had a case in which there was calcification of the aortic valve, and Streptococcus zymogenes was recovered from the calcified material. Dr. Libman examined the material and regarded the changes as those of healing subacute bacterial endocarditis. In the cases originally reported by Dr. Libman, Dr. Baehr went over the sections of kidney and found evidence of a healed focal embolic glomerulonephritis. I wonder whether Dr. Richter observed anything in the kidney that might be regarded as a manifestation of a healed focal lesion.

MAURICE N. RICHTER: In regard to healed or healing subacute bacterial endocarditis, I have had no experience.

As far as calcification resulting from infection is concerned, I think from a study of the sections that the calcium is deposited in the necrotic muscle fibers rather than in abscesses, so that I should hardly regard this as healing of a focal infection. There was no evidence of focal or embolic glomerulonephritis, and the only embolic phenomenon observed at autopsy was infarction of the spleen. There were, during the last few days of life, clinical evidences of cerebral damage, but no permission to examine the brain could be obtained.

NEPHROSIS DUE TO CARBON TETRACHLORIDE POISONING. HANS SMETANA (by invitation).

CASE 1.—A 71 year old white man, a photographer, drank a glass of carbon tetrachloride by mistake. He was subject to chronic alcoholism. Shortly after having taken the chemical he became nauseated and vomited persistently. Jaundice developed on the following day and persisted. On the sixth day he became anuric; ascites developed, and the patient died in uremic coma after ten days of illness. The temperature was 97.4 F.; the pulse rate 80; the respiratory rate 22; the blood pressure 140 systolic and 85 diastolic on the eighth day of illness.

Laboratory Findings.—The hemoglobin content was 120 per cent of normal; the red blood cell count, 5,600,000; the white blood cell count, 13,680, with 91 per cent polymorphonuclears. The serum inorganic phosphorus was 7.8 mg. per hundred cubic centimeters; the calcium, 8.7 mg.; the nonprotein nitrogen, 165 mg.; the phosphatase, 6.3 Bodansky units per hundred cubic centimeters; the serum protein, 7.2, and albumin, 3.7 per cent. The urine showed albumin (4 plus), bile (2 plus), red and white blood cells and casts.

The autopsy showed extensive central necroses in the liver and, in the kidneys, nephrosis, characterized by distention of glomerular spaces with albuminous precipitate, swelling and vacuolation of epithelial cells of the proximal convoluted tubules, degeneration and necrosis of cells of the distal convoluted tubules and loops of Henle, and hyaline, granular and cellular casts in the lumens of the tubules. In addition, highly refractive round concretions were present in the lumens of the tubules.

CASE 2.—A 37 year old colored housemaid with a history of addiction to alcoholic beverages, became ill after having cleaned dresses with carbon tetrachloride in a close, stuffy room for three hours. After having finished her work she drank a glass of milk. She was nauseated, vomited repeatedly and began to have diarrhea. On the following day signs of tetany developed, with Chvostek's and Trousseau's signs. Six days after her exposure to the fumes of carbon tetrachloride, she became anuric in spite of large infusions of Ringer's solution and dextrose. There was tenderness in the region of the liver, but no jaundice. The temperature was 103.6 F.; the pulse rate, 88; the respiratory rate, 26, and the blood pressure, 130 systolic and 78 diastolic.

Laboratory Data.—The hemoglobin content was 85 per cent; the red blood cell count, 4,300,000; the white blood cell count 13,250, with 87 per cent polymorphonuclears. The serum inorganic phosphorus was 5.6 mg. per hundred cubic centimeters; calcium, 8.1 mg.; phosphatase, 4.7 Bodansky units; bilirubin, direct, 4.7 mg.; nonprotein nitrogen, 94 mg.; total plasma lipids, 1,185 mg.; neutral fats, 760 mg.; serum protein, 7.8 per cent; albumin, 4.4 per cent. The urine contained albumin (4 plus); bile (1 plus), white and red blood cells and casts.

After nine days of illness the patient's output of urine rose to over 1,000 cc. daily, but the nonprotein nitrogen continued to be high for some time. She then had lobular pneumonia, with myocardial damage, but recovered uneventfully and was discharged after forty-six days of illness.

CASE 3.—A 35 year old colored dry cleaner with a history of alcoholism became acutely ill with headache, dizziness and malaise after having cleaned furniture with carbon tetrachloride for several hours. He was dyspneic, began to have a

cough, became nauseated and vomited repeatedly. He became jaundiced and anuric on the sixth day of his illness and died in uremic coma eight days after having been exposed to carbon tetrachloride. The temperature was 99.4 F.; the pulse rate, 130; the respiratory rate, 36, and the blood pressure 220 systolic and 125 diastolic.

Laboratory Findings.—The hemoglobin content was 97 per cent; the red blood cell count, 5,700,000; the white blood cell count 36,000, with 95 per cent polymorphonuclears. The nonprotein nitrogen of the blood was 188 mg. per hundred cubic centimeters. The venous pressure was 165.

The autopsy showed petechial hemorrhages in all serous membranes and in the urinary passages and large hemorrhages in the mesentery and retroperitoneal tissues. The lungs showed edema and early consolidation; the liver was fatty, with extensive central necroses. The kidneys were greatly swollen and microscopically presented nephrosis identical in character but more pronounced than that described in case 1.

The clinical features common to all the cases were fever, nausea, vomiting, jaundice, anuria, retention of nitrogen, hypertension, polycythemia, leukocytosis, and albumin, red and white blood cells, bile and casts in the urine. Additional features were tetany in case 2, hemorrhagic diathesis in case 3 and involvement of the lungs in cases 2 and 3. All the victims were chronically addicted to the use of alcohol. The anatomic observations in the two fatal cases were essentially the same and differed only in degree, hepatic damage being more extensive in case 1 and the renal lesions more pronounced in case 3. This was probably due to the difference in mode of intoxication.

The anatomic basis found for the renal symptoms in all the cases was nephrosis, characterized by swelling and vacuolation of the proximal convoluted tubules, degeneration and necrosis of cells of the distal convoluted tubules and loops of Henle, and the presence of granular, hyaline and cellular casts in the tubules and the space of Bowman.

DISCUSSION

IRVING GRAEF: Through the courtesy of the Chief Medical Examiner's Office I have had the opportunity of studying the kidneys and other organs in 2 fatal cases of carbon tetrachloride intoxication in the last year at Bellevue Hospital. In 1 case the ingestion of carbon tetrachloride occurred at a drinking party in which the participants ran out of gin and substituted carbon tetrachloride. The other case was that of a man who had been drinking for several days in a small room with a drinking partner, and while he was intoxicated a cigaret which he was smoking ignited his mattress. A fire ensued, which was put out by the use of a fire extinguisher, the latter being sprayed generally around the fire, the mattress and the occupant of the bed. Both patients began to show signs of acute yellow atrophy and died about a week after the ingestion or inhalation of the carbon tetrachloride. I was much impressed by the extent of the hepatic lesions in these cases. The lesions were quite like those which Dr. Smetana has showed, and the renal lesions resembled those he described, but I have been hard put to it to classify the renal lesions. The kidneys reminded me more of those one sees after hemolytic reactions when a transfusion has been done with incompatible blood. While there was evidence of some tubular damage in both these cases, I did not see the severe necrosis of the tubular epithelium that one ordinarily associates with necrotizing lesions. But there was widespread obstruction by intratubular casts. Some of these seemed to be composed of red cells, and I wonder whether hemolysis was taking place in these patients. I noticed that Dr. Smetana's clinical reports refer to hematuria: It seems to me possible that what passes for hematuria on ordinary microscopic examination may represent a reprecipitation of laked blood in the form of droplets or globules, such as one sees in the kidney after a hemolytic reaction. The role of the renal lesion in causing death has not been evaluated yet, to my knowledge, and I am not quite sure that I know whether Dr. Smetana means that the renal lesion is responsible for death in these cases.

One other point is pertinent here. Perhaps the members of the society have not come across a rather important experimental report by Neale, of the Franklin Institute, Philadelphia, who has protected rats from hepatic damage by the use of sodium xanthine and sodium guanine, given parenterally in rather small doses. These give rats protection against the fatal effect of carbon tetrachloride after periods of deep anesthesia for one hour and forty minutes. The members of the society might like to know about this report (Neale, R. C.: *Science* **86**:83, 1937) because of the importance of carbon tetrachloride as a cause of acute yellow atrophy.

HANS SMETANA: In what fluid were the kidneys fixed?

IRVING GRAEF: Solution of formaldehyde U. S. P.

HANS SMETANA: It may be that these precipitates resemble blood in formaldehyde-fixed preparations. The material which I studied was fixed in Zenker's solution. In the cases reported there was never gross hematuria but only a few red cells in the urine. Blood was looked for in the tubules very carefully. The peculiar granular material looks like precipitated protein, not like hemoglobin casts.

Whether death is due to the renal lesions is difficult to say. In some cases apparently the damage to the liver is more extensive, while in other cases the kidneys are more affected, depending perhaps on the mode of intoxication. Thus in case 1 the most important lesion was the acute yellow atrophy, while in the second and third cases the renal lesions were more extensive. In all 3 cases the clinical picture was that of nephrosis. Chandler and Chopra (*Indian M. Gaz.* **60**:406, 1925), who reported experiments with carbon tetrachloride on cats, attributed the death of their animals to the renal lesions rather than to the hepatic damage.

As far as the reference to guanidine is concerned, I have not read the paper mentioned by Dr. Graef. Carbon tetrachloride poisoning has been compared to guanidine poisoning by Minot and Cutler (*J. Clin. Investigation* **6**:369, 1928) because of the increase of guanidine in the blood in experimental carbon tetrachloride poisoning. On the basis of their work carbon tetrachloride poisoning is treated with calcium carbonate or lactate.

PROGNOSTIC SIGNIFICANCE OF INTRACELLULAR MUCICARMINOPHILIC MATERIAL IN CARCINOMA OF THE FEMALE BREAST. VIRGINIA KNEELAND FRANTZ.

Delbet and Mendaro (Les cancers du sein, Paris, Masson & Cie, 1927) published the statement that "secreting" tumors are relatively more benign than "nonsecreting." This statement was based on preparations stained with mucicarmine. Six other papers have since appeared on the subject of this particular stain. Most of the series were small, and in none was the follow-up extensive.

A study is here presented of 157 consecutive cases in which radical mastectomy was done at the Presbyterian Hospital from May 1928 through May 1932, with a follow-up study that included 97.1 per cent of the cases. Twenty-seven cases had to be eliminated for various reasons, leaving for analysis 130 cases, all of which were followed up.

These tumors were then graded according to the plan submitted by Haagensen in 1933. The grades were compared with the results of the subsequent investigations, which covered a minimum of five and a maximum of nine years. Sixty per cent of the patients had axillary metastases at the time of operation, as demonstrated in the laboratory. The figures are based on the number surviving for five years *free from evidence of disease*. Good results were obtained in 35.3 per cent of the group as a whole, those without axillary metastases showing 61.6 per cent good results and those with axillary metastases 18 per cent. The figures for five year survivors are naturally somewhat higher in all groups. It is obvious that the presence of metastases far outweighs in prognostic significance any histologic character.

The presence or absence of intracellular mucicarminophilic material was then determined. No interpretation of the nature of this material was made, though

there is suggestive evidence that it is a true secretion and not a degeneration product. Figures are given to show that the material was found in tumors of all grades but in decreasing amounts as the grade of malignancy increased.

The cases were then analyzed according to the follow-up and the presence or absence of the material mentioned and further subdivided according to the presence or absence of axillary metastases. In the group in which the material was not present there were 29.2 per cent good results, and in the group in which it was present there were 39 per cent good results. In the most significant group, that without demonstrated axillary metastases, there were 46.1 per cent good results in the tumors in which the material was not present and 76.9 per cent good results in those in which it was present.

The cases were further studied according to the dates of death and the number of patients living each year, and in both these analyses the persons whose tumors contained the intracellular mucicarminophilic material showed better results. It was therefore concluded that the presence of the material was a moderately favorable sign—one, however, which had to be interpreted in the light of other histologic criteria.

The histologic criteria for grade 1 and and for grade 3 are so striking that information about this material is not needed in determining the grade. Grade 2, however, which constitutes the largest group, is made up of those cases which are not frankly of grade 1 or grade 3. It is thought that study with the mucicarmine stain may be of assistance in determining the relative malignancy of tumors of this group. Analyzed separately, the tumors in which the material was present showed 72.2 per cent good results, and those from which it was absent, 11.1 per cent good results.

It is again emphasized that grading of tumors offers only minor additional information and that the outcome in any given case depends on other factors, often not determinable, such as duration and extent of disease.

DISCUSSION

N. CHANDLER FOOT: I should like to ask Dr. Frantz a rather pointed question: What amount of demand is there on the part of surgeons for grading of tumors and what amount of reliance does she place on the grades given out? I have had some doubt about the value of grading tumors from the standpoint of the variety of grades one may see in a given tumor if one takes different parts of it, and also from the standpoint of the dangers inherent in the method. If the grades are not determined with extreme care and if they should be handed out by men who determined them superficially (and I am sorry to say that the determinations are often done just "by guess and by gracious") they would have little value.

ALFRED PLAUT: I think I can come to the aid of the American literature by stating that Dr. Robert Greenough, of Boston, about twelve years ago wrote a paper on the prognosis of breast cancer in relation to secretion.

I should like to ask Dr. Frantz whether her work was never disturbed by the presence of glycogen, which also stains beautifully with mucicarmine. What Dr. Frantz has shown obviously cannot be glycogen, because of the way it is situated in the cells and tissue.

I might add a word to what Dr. Foot has said about the value of grading. Years ago I was made to busy myself with the histologic prognosis of tumors, and in general my reaction to it today is as negative as then. I am appreciative of the careful way in which Dr. Frantz has weighed all the single anatomic considerations. A great difficulty lies in the fact that when one tries to analyze carefully, the material will break up into groups so small that statistical conclusions become impossible.

VIRGINIA KNEELAND FRANTZ: In answer to Dr. Foot, I must acknowledge that my associates and I never tell the surgeons what the grade of a tumor is, because we fear that they will place more reliance on it than they should. We do feel that it is an advantage to arrive at some estimate of the relative malignancy

of the cancers with which one is dealing. Moreover, with the advent of the enthusiasm, particularly at the Memorial Hospital, for radiotherapy in cases of cancer of the breast, it seems as though the further we can study the histologic detail that may be of prognostic significance, the more reliance we may put on it. But we are well aware that a cancer which we may classify as grade 1 tomorrow may end fatally within six months, even though no axillary metastases are demonstrable at the time of operation. There is one such in this series. The patient was dead in eleven months with pleural metastases, yet no axillary metastases were found in the operative specimen. Distant metastases must, of course, have been present. We place little reliance on grades, and we do not emphasize them in our reports, but when the French authors came forward with this differential staining, the staining was easy to do, and we had a hope that there might be something in it.

As to the variability of the grade because of the variability of the tumor, we have made it a rule in evaluating any tumor to lay emphasis on the most malignant portion of the tumor. For instance, we consider a tumor as a "comedo" carcinoma only if we find that all the tumor is confined to the ducts. If there are portions of the tumor outside the ducts and it is of an anaplastic, infiltrating type, we should not consider that as a "comedo" carcinoma but would take the most malignant portion as significant.

I am aware that in tissues prepared with the mucicarmine stain glycogen might be confused with mucin. I believe that there would not be a great deal of glycogen in these tumors. I have asked biochemists about that. In their analyses of tissue from different parts of the body they have found that the breast has not a great deal of glycogen. With eosin-methylene blue, which stains mucin a faint greenish tinge in those cells in which it is considered true secreted mucin, this intracellular material has a greenish tinge. I have no way of knowing whether this was all mucin or a mixture of substances which ended in being pink in the stain. I am inclined to believe it represents secretion and that therefore it may be regarded as an indication of function of the cell, but I have no way of proving this. It is a striking finding, which we thought it might be well to investigate with a view to determining its significance.

I also am well aware that such a series breaks down into small groups so that the figures mean little. However, in all of the grades and in all of the groups, with or without axillary metastases, the figures fell in this series on the same side of the line, although the spread of the curves was not wide.

NEW YORK PATHOLOGICAL SOCIETY AND NEW YORK GASTROENTEROLOGICAL ASSOCIATION

ALFRED PLAUT, *President, New York Pathological Society, in the Chair*

Joint Meeting, March 29, 1938

EDWARD L. KELLOGG, *President, New York Gastroenterological Association*

ROBERT A. MOORE, *Secretary, New York Pathological Society*

GEORGE E. BINKLEY, *Secretary, New York Gastroenterological Association*

HEREDITY IN CANCER. MADGE THURLOW MACKLIN, London, Ontario, Canada (by invitation).

The medical profession begins to appreciate the hereditary basis of cancer as it becomes aware of (1) the laws of heredity, (2) the distinction between hereditary and congenital conditions and (3) the biologic significance of the fact

that cancers are inherited in insects, fish and mammals. Although the details of hereditary transmission in one form cannot be applied to another, the principle that genetic factors are a basis of cancer will be true in all forms.

Although cancer occurs frequently in the population, familial incidence of cancer, especially that in which 4 or 5 members of a family are affected, would not be found as often as it is if cancer were not hereditary. This is especially apparent when it is considered that the *capacity for developing a specific type of tumor at a fairly definite time* seems to be the factor that is inherited. One can calculate in how many of 3 or 4 sisters mammary or ovarian carcinoma should develop on the basis of chance and find this occurrence much oftener than chance allows for. Husbands and wives, who are usually unrelated but have a similar environment, have cancer of the same type, such as gastric, only one-tenth as often as related members of a family, such as parent and child or children in a family who have similar heredity in part, as well as similar environment.

Identical twins, i. e., twins with totally similar heredity, are both affected with the same type of tumor at nearly the same age much more often than twins with a totally dissimilar heredity, i. e., nonidentical twins. Therefore tumors have a genetic basis, for the following three reasons: 1. Related members of a family have the same type of tumor in far too many instances to be accounted for on the basis of chance distribution. 2. Members of a family not related, e. g., husband and wife, are not both affected in nearly so many cases as related members of a family, although sharing a similar environment. 3. Twins with an identical heredity having the same type of tumor are observed in much larger numbers than are twins with the same type of tumor in whom the heredity may or may not be identical.

Acceptance of genetic factors as the basis of cancer does not preclude acceptance of external factors as modifying or aiding the genetic ones. I believe that all persons inherit the capacity for cancerous change in their tissues. Whether one gets cancer or not depends on whether that one lives long enough for his "cancer alarm clock" to ring. If it is set to go off at 100 years, then for all practical purposes that person is cancer immune. If it is set to go off at 60, he may live to see cancer develop. Some irritant may move the hands of the clock forward, so that the alarm rings twenty or thirty years before it would have without the irritant. The type of tumor that develops depends on the specific organ for which the earliest alarm was set, as there may be present capacities for the development of several types of tumor. Here again external factors may modify the genetic picture, roentgen ray dermatitis turning into cancer at 50 when gastric cancer is not set to develop until 70.

The medical profession is aware of the relation of "sex hormones" to tumor development. It is known that families differ from others chemically in that some manufacture pentose or levulose instead of dextrose; others show the development of alkaptonuria or cystinuria. All persons differ from all others in the chemical processes of their sweat or sebaceous glands, just as all persons differ in their fingerprints; otherwise dogs would not be able to trace their masters by scent. May it not be that the hormones of different persons differ chemically, some being carcinogenic as well as developing sex characters and others being noncarcinogenic? It is known that a shift in position on the benzene radical of a methyl group alters the carcinogenic properties of a compound. Bile acid metabolism in some may produce carcinogenic end products; in others, not. This is a fertile field for research.

Finally, it is not possible to breed cancer out of the human race, no matter what the mode of inheritance and no matter how good the records that are kept, because it develops too late in life, i. e., when the reproductive period is almost or wholly over. It would be necessary to breed it out of the entire race, as well as the good qualities that are often found in persons who have cancer. Different tumors exhibit different types of inheritance, and the same tumor may exhibit different types of inheritance in different families. Thus laryngeal cancer is

infrequently met with in 2 in a family as compared with rectal, gastric, breast or uterine cancer. Because no one knows the mode of the transmission of cancer, and because cancer occurs too late, one cannot eliminate cancer by breeding it out.

EXPERIMENTAL EVIDENCE ON THE RELATION BETWEEN HEREDITY AND EXTERNAL FACTORS IN CANCER. CLARA J. LYNCH (by invitation).

At present most investigators in this field believe that the manifestation of neoplastic disease is the result of both heredity and environment, and further, that the proportion of these two factors varies for different tumors. The present paper traces the influence of intrinsic and extrinsic factors in tumors of the lungs in mice, leukemia, cysticercus sarcoma in rats and mammary tumor in mice. It also takes up certain considerations that have a bearing on the problem of cancer as a whole.

It has been shown both by the existence of strain differences among mice and also by breeding tests that susceptibility to spontaneous tumor of the lung is inherited. Pulmonary tumors may be induced by tar, and with this technic the influence of environmental change and of heredity has been clearly demonstrated. A comparison of the incidence of tumor induced simultaneously in two different organs of the same individuals shows that the susceptibility to tumor is organ specific. Therefore, cancer is not due to one hereditary unit; there must be separate mendelian genes to govern the susceptibility of different organs. MacDowell and Richter showed both by strain differences and test crosses that heredity controls susceptibility to mouse leukemia but that variability due to undetermined environmental forces is operative. In the cysticercus sarcoma of rats, in which the parasite represents the extrinsic factor, Curtis, Dunning and Bullock concluded that heredity is important only so far as it affects longevity and susceptibility to cysticercus disease. They extended their theory to include other tumors and suggested that the inherited character always consists in the control by the individual of the amount of the incitant and not in differences in tissue susceptibility. In the study of the mammary tumor of mice, though strain differences show the influence of heredity, the physiologic state of the individual influences tumor incidence. Ovariectomy, breeding, intensive breeding with prevention of nursing and injections of an estrogen affect the incidence. It has not been shown whether an estrogen is directly carcinogenic. Bagg suggested stasis in the mammary gland as the tumor incitant. Certain authors (Loeb, Lacassagne and others) believe the inherited character is a sensitivity of the tissues to an estrogen and possibly other hormones rather than degree of control over the amount of estrogen produced or of the rate of its elimination from the body. Former calculations as to the number of genes involved in the inheritance of susceptibility to mammary tumor have been upset by the discovery of maternal influence (Little and co-workers; Murray, Korteweg and others). Bittner's data indicate that something may be present in the milk taken by suckling young which affects the genesis of mammary tumor. Murray concluded that in a certain experiment the evidence from F_2 virgin females showed the extrachromosomal influence to be six times stronger than the chromosomal. It may be that under other conditions this proportion will not hold. Inherited differences in constitution are still basic for the manifestation of mammary tumor. MacDowell and Richter found that leukemia and longevity were affected by maternal influence, but that fostering the young had no effect. The question of somatic mutation as the cause of malignant change must remain unanswered until new methods for the solution of the problem are developed.

DISCUSSION

MAURICE N. RICHTER: The papers that have been read on the influence of heredity and environment on cancer offer a number of interesting phases for discussion. It is of interest that they deal with different types of material

analyzed in different ways; yet the authors have reached the same conclusion, namely, that heredity is important in the development of cancer and that external influences are also important. It is also interesting that this conclusion has been reached by practically all geneticists who have worked with cancer. Geneticists agree pretty well among themselves that cancer is under the influence of heredity, although they disagree on minor points, such as how heredity acts in bringing about this change. One difficulty in dealing with cancer is that no one really knows what cancer is. If one is going to discuss cancer, one ought to start with definition, and of these, one may find a great variety. The clinician defines cancer in terms of what it does in the body and to the body; the pathologist defines it in terms of its gross and microscopic appearance; the chemist defines it in terms of chemical changes in the body or in the tumor, and the geneticist probably brings in hereditary factors.

I think Dr. Macklin is working under difficulties in dealing only with the human race. Geneticists deal mainly with pedigrees, with data that can be subjected to statistical analysis, and the data on cancer in the human race are meager. The statistics do not go back far enough. The recognition of cancer was inaccurate even no farther back than one or two generations. There are no accurate figures on the occurrence of cancer in our ancestors. Dr. Macklin has shown remarkable ingenuity in using these meager statistics to the best advantage. Unfortunately there is one other feature of the study of human cancer which Dr. Macklin pointed out, and that is the inability to perform genetic tests, which to the geneticist are of prime importance in all experimental work. In animals it is possible to obtain strains that are genetically uniform. It is possible to mate one strain with another and by analysis of the results obtain information that cannot be obtained in the human race. For that reason all work with human cancer is at a disadvantage.

If heredity were the only factor of importance in the development of cancer, the clinicians would also be at a great disadvantage, for the only way in which they could lower the death rate from cancer would be to permit people to die at an early age from infectious disease or like causes. Little else could be done about it, although knowledge of the patient's ancestry might change the clinician's point of view in other ways. If it is known that in a certain family cancer of a certain organ is likely to develop, the clinician may be on the lookout for it and recognize it while it is still in a stage at which therapy can be instituted.

However, if it is shown that external influences also affect the incidence of cancer, there is an opportunity of studying the disease through these external influences; there is also in this the best opportunity of preventing and curing the disease by control of these influences.

Not all of the external factors that may influence the occurrence of cancer are known, but their importance cannot be overemphasized. As Dr. Lynch mentioned, they are extremely important in experimental work. If both heredity and environment are of importance in the development of a characteristic, and if a study is to be made of their relative importance in the formation of this characteristic, it is necessary to control one or the other factor, for the experimental method implies that all factors except the one studied are controlled. It is easy to control temperature, time, age, sex and other such factors in experimental work. It is also easy, given the time and the patience, to control heredity; but this is the one thing that has been controlled least of all in most experimental work. It has been shown by mathematical calculation that if one starts with 2 separate individuals, mates them and in the offspring continually mates brother to sister in each generation, no matter how diverse the characters of the original 2 parents may have been, the characteristics of the offspring will become more and more uniform until one finally obtains a strain of animals which are genetically uniform. Eleven generations of such inbreeding, according

to the figures of Wright, will give a degree of genetic uniformity of 95 per cent. Only when a stage of uniformity in the genetic constitution is reached can one study nongenetic factors, because only then can one keep the genetic factor uniform.

There is one other feature of cancer that has not been mentioned. I think it worth mentioning because it shows the influence of heredity and environment in a somewhat different way. The discussion has been concerned so far with the influence of heredity and environment on the spontaneous development of cancer cells in a body. Just how this development takes place is not known. It may be a somatic mutation; it may be something else. Nevertheless, the change must occur, and no one knows how often this change occurs in any particular individual, because only those changes are observed that can endure. Certain cells may undergo a change that will convert them into cancer cells, and yet if the body in which they are growing is not suitable for the growth and development of cancer cells, they will not grow, and the patient will not have cancer, even though cancer cells have arisen. A similar observation may be made experimentally by removing cancer cells from the body in which they arose and injecting them into other animals. It is then observed that the genetic constitution of the host is one factor that determines whether the cancer cells will survive in their new environment. My own experience in this field has been limited to leukemia in mice, but leukemia in mice behaves, as far as the features under discussion are concerned, much like other transplantable tumors in mice. One can remove the cells, the leukemic cells, from a leukemic mouse and inject them into other mice, but whether or not leukemia will develop in the other mice after such inoculation depends in part on hereditary factors and in part on nonhereditary factors. As an example, in the strain of mice that Dr. Lynch mentioned, 90 per cent of the mice, if they live more than six months, will show spontaneous development of leukemia. It has been shown that the high incidence of leukemia in this strain is due to heredity. One can take leukemic tissue from any one of these mice and inject it into any other mouse of the same strain, and these other mice will show the development of leukemia in 100 per cent of the cases, and by continuous transference from one mouse to another a line of leukemic cells can be kept going indefinitely through one mouse after another. If, however, one takes the same cells and injects them into mice of a different, unrelated strain, leukemia may or may not develop, according to the genetic constitution of the mice into which the cells have been inoculated. My co-workers and I have a strain in which over long periods of time it has been practically impossible to induce leukemic cells to grow. By mating these two strains, one of which is susceptible, one of which is not, we can follow by genetic analysis the hereditary influence on inoculation. But susceptibility is not only under the influence of heredity; it can be influenced by other factors; for example, in animals which are 100 per cent susceptible to inoculation of leukemic cells, immunity can be developed by injecting certain tissues before the inoculation of the leukemic cells. One can do this by injecting tissues of normal mice, provided these mice have a genetic constitution which is different from that of the susceptible strain. One can do it also by injecting embryonic tissue, and again the genetic constitution of the embryonic tissue will influence the development of leukemia in these animals. These results with leukemia are comparable in many ways to those obtained by other workers who have used the inoculation method of studying experimental tumors.

ALFRED PLAUT: It has been accepted by many that cervical lacerations from childbirth are one of the main predisposing factors in the development of carcinoma of the cervix. May I venture to state that no statistical material of any real value is available to prove that point? I do not believe that there is any close connection between laceration of the cervix and the development of cervical carcinoma. I do not know why, with such abundant material available, there has been no convincing statistical study of this important problem.

MADGE THURLOW MACKLIN: The statement of Dr. Richter that human material cannot be experimented with is of course a great tragedy for any one who is trying to study human material. One cannot have back crosses, and one cannot mate brother and sister. If one did, all the social welfare departments in the state would be at one's heels for ruining the morals of the community. No one regrets this inability to check conclusions more than the person who studies human records. Nevetheless, I do feel that although my colleagues and I do not have the long pedigrees that Dr. Richter has for mice, and although we cannot go back more than one generation or so and get a definite diagnosis, the very fact that we can frequently get data showing that at least two or more persons in a family have a very rare type of tumor, the diagnosis of which is confirmed, is something which may be as significant from the statistical standpoint as a long pedigree showing heredity of a much commoner character. I remember asking Dr. Fisher: "What is the statistical significance if a pair of identical twins have the same tumor at the same age?" His reply was, "I may say there is not much to say against heredity there." I went on to ask: "If you could produce records of 10 or 25 or 50 such pairs of identical twins with the same tumors, what would be the conclusion?" And he said, "You need not go any farther, because by the time you have ten pairs you have enough to prove the point absolutely." So I do not think any one has to worry much about the lack of long pedigrees in the human race.

There is one thing which I think is of value in the study of the inheritance of tumors: Not only can one study these cases early because some one else in the family has a tumor, and treat the subjects before they even have symptoms of the tumor, but one has the opportunity of finding out the potentially susceptible members. In Russia investigators of the problem have been doing that in the case of gastric cancer; they have been analyzing gastric contents in persons belonging to families who show gastric cancer, and they have found a much higher incidence of achlorhydria in such people than in the population at large. Whether these achlorhydric persons may be potentially subjects of gastric cancer or not, the Russian investigators do not know, but a follow-up system will probably reveal the percentage that are conditioned for gastric cancer so that the predisposition can be treated earlier, without waiting for symptoms. Hence I feel that the study of the inheritance of tumors in man has some value.

CLARA LYNCH: I am glad that Dr. Richter brought up the point of the transmission of leukemia and of transplanted tumor. The evidence for the inheritance of susceptibility to transplanted tumor is very clear indeed. A number of these cases have been analyzed, and it is possible to determine the actual number of genes involved. The transplantation of tumor really is like the transplantation of any other kind of tissue; it is not so much cancer that is being studied as the laws underlying transplantation in general.

There is one point in regard to susceptibility to stomach and intestinal tumors which I should like to touch on. Little experimental work has been done on these tumors in mice. Tumors of the stomach and intestine have been reported; occasionally they were in animals that were somewhat related, but as a rule they have been rather scattered in the pedigrees. I have one pedigree which shows the descendants of a mouse which was found at autopsy to have carcinoma of the stomach. There were about 150 descendants in the first and second generations. Carcinoma of the stomach, or at least tumor (it was apparently not malignant), appeared in only 1 of these mice, so that the evidence for heredity was not striking.

I am going to encroach on Dr. Macklin's field in regard to the evidence to be derived from identical twins. I have found in the literature 8 pairs of one ovum twins with tumor of the gastrointestinal tract. In 4 of these pairs only 1 twin per pair had carcinoma of the stomach or intestine. There were no pairs in which both twins had tumors of different organs. In 4 of the pairs both twins had tumors of the same type affecting the same organ—that is, if

you will admit that the pylorus and the rectum are the same organ. Of dizygotic twins, there were 13 pairs. Seven of these pairs had tumor of 1 twin per pair. In 5 pairs both twins in each pair had tumor but of different organs. There was only 1 pair in which the 2 twins had tumor of the same organ. Admittedly, these numbers are small. They are consistent, nevertheless, with the idea that susceptibility to cancer of the stomach or intestine is hereditary. They show, however, that heredity is not the only factor. There is a case in which 1 twin had carcinoma at the age of 46 and the other has lived twenty-one years longer. At the latest report, he was still alive at the age of 67 without having had a tumor of any sort.

CHICAGO PATHOLOGICAL SOCIETY

FRANCIS D. GUNN, *President*

Regular Monthly Meeting, March 14, 1938

EDWIN F. HIRSCH, *Secretary*

EXPERIMENTAL TUMORS OF THE BRAIN IN THE WHITE RAT. ARTHUR WEIL.

The report will be published in full in a later issue of the ARCHIVES OF PATHOLOGY.

DISCUSSION

S. ROSENTHAL: Usually squamous cell carcinomas of the brain are considered to be teratomas.

V. LEVINE: Where do you think the squamous epithelium originated? Were the cells carried in during the injection or were they from epithelial structures at the base of the brain?

ARTHUR WEIL: The epithelial cells were not carried in with the needle, but probably represented the ependymal lining with neoplastic evolution into tumor.

P. BAILEY: There are many reports of papillary epithelial tumors of the third ventricle, and ependymal cells seem to transform into squamous cells. However, a silver stain always demonstrates a basement membrane. The relation of these cells to the underlying neuroglia should be determined.

EXPERIMENTS WITH THE GORDON ENCEPHALITOGENIC AGENT IN THE STUDY OF HODGKIN'S DISEASE. PAUL E. STEINER.

The most interesting recent contribution to the etiology of Hodgkin's disease has been the discovery of the Gordon encephalitogenic agent. The presence of the agent in the enlarged lymph nodes has been made the basis of a diagnostic test, since this active principle is absent from the lymph nodes in other lymphatic diseases. Gordon (Rose Research on Lymphadenoma, Bristol, John Wright & Sons, Ltd., 1932) discovered that the diseased lymph nodes of patients with Hodgkin's disease contained an agent which was able to produce a characteristic syndrome of spastic paralysis, ataxia and other symptoms on intracerebral injection into rabbits and guinea pigs. He favored the opinion that the agent is a virus and published photographs of "elementary bodies" in the active preparations (*Proc. Roy. Soc. Med.* **27**:1035, 1934).

As a diagnostic test I have found the Gordon reaction reliable. Of 14 cases of Hodgkin's disease in which the diagnosis was made by histologic examination, the test has been positive in 12. Considering that two variables are in comparison, this correlation is excellent. In 22 cases of other lymphatic diseases, including

lymphosarcoma, tuberculous lymphadenitis, nonspecific lymphadenitis, aleukemic leukemia and leukemia of monocytic, myelogenous and lymphatic types, studied as controls, the results were negative. My results agree with the experience of others.

The test does not replace but supplements histologic examination, just as the inoculation of guinea pigs is used in diagnosing tuberculosis. The greatest usefulness is in cases in which the histologic observations are atypical or inconclusive; in such cases it reduces the necessity for repeated biopsies. Since an incorrect diagnosis of Hodgkin's disease can have serious consequences, this is a useful addition to the diagnostic tests of the laboratory. The lymph node suspension can be made quickly at the time of biopsy, and then, if the microscopic examination is inconclusive, the animal test can be made.

The active agent has been considered a virus by some, a neurotoxin by others and a proteolytic enzyme by still others. It has some attributes of each of these agents. Macerated tissues in 10 per cent suspension in nutrient broth show an increase in activity during several weeks, indicating that the active principle either is increasing or is being released from cells by autolysis. The agent has survived for as long as three months, even in the presence of 0.5 per cent phenol. It has not survived for four months. Injected into the spinal canal in the lumbar region, it causes flaccid paralysis of the hindlegs, but there is no ascending infection. Injected into the sciatic nerve, it does not ascend to the spinal cord. Passage experiments with the brains of recently diseased and of chronically diseased rabbits have given uniformly negative results. Others have reported that the agent is filtrable, that immunity is not conferred by one attack, that the agent survives heating to 60 C. for one-half hour, freezing to 190 C., desiccation and irradiation and that it is destroyed by acids and alkalis.

A closely related, if not identical, agent has been reported in normal and diseased (pernicious anemia and leukemia) bone marrow of man and in monkey's marrow and spleen, as well as in a few other tissues. I have found it present in normal human bone marrow but absent in three spleens in which the infiltration characteristic of Hodgkin's disease was marked. The buffy coat from blood has not shown it. It does not appear to be in the lymph nodes of patients with Hodgkin's disease as the result of the presence of the diphtheroids commonly found, for filtrates from two such strains failed to give the reaction. The presence of the agent elsewhere than in lymph nodes does not reduce its usefulness as a diagnostic test for lymphomatoid diseases.

Correlation of the histologic structure of the lymph nodes and the degree of activity has not impressed me or others (Chapman, E. M.: *Ann. Int. Med.* **10**:742, 1936. Van Rooyen, C. E.: *Edinburgh M. J.* **44**:455, 1937). Recently Turner, Jackson and Parker (*Am. J. M. Sc.* **195**:27, 1938) attributed the activity to the presence of eosinophils.

Although the chief usefulness of this test at present is as an aid in diagnosis, it has great potential value for the elucidation of the conditions that are called "atypical Hodgkin's disease." If, as many believe, a number of diseases of different etiologic backgrounds, such as obscure fungous diseases and reticulum cell lymphosarcoma, are masquerading as, and are diagnosed as, Hodgkin's disease, or if the cellular reaction which at present is diagnosed as Hodgkin's disease represents the body's reaction to a variety of etiologic agents, this test may prove of value in arriving at an etiologic classification. Should Gordon's hypothesis that the active principle is a virus be substantiated, the possibility of finding a curative agent for this disease is increased.

MORPHOLOGIC VARIATIONS OF TUMOR CELLS. OTTO SAPHIR.

A study of miscellaneous carcinomas revealed morphologic variations among the individual tumor cells of particular neoplasms. These morphologic differences were not those always encountered in malignant tumors but were often so hetero-

geneous as to obscure the exact nature of the tumor. They appeared principally to reflect the presence of seemingly different types of tumor cells in the individual tumor. Thus, basal cells or transitional cells were found in squamous cell carcinomas.

The presence of various types of tumor cells in the same tumor may be due to a number of factors. However, the results here presented seem to indicate that these tumors arise not from individual cells or from certain cell groups but from one area which contained the various cells of which the tumor consists or derivatives of these cells.

Morphologic variations must be taken into consideration in the grading of carcinomas and also in the determination of the sensitivity of the tumors to radioactivity. The relative resistance of some basal cell or transitional cell carcinomas to radium could be ascribed to the presence of many squamous cells in both of these tumors.

DISCUSSION

PAUL E. STEINER: In a discussion of the causes of such tumors a possible genetic basis should be considered. Dr. Slye has recently demonstrated in her tumor mice, a factor for localization of tumor in addition to the factor for type of tumor. She has reported strains of mice which have mixed carcinosarcomas of the mammary glands because factors for both carcinoma and sarcoma are present. Morphologic variations may depend on what genetic potentials are in the cells.

THE HISTOLOGIC PICTURE OF VASCULAR SPASM IN EXPERIMENTAL GASTRIC ULCER. ALEXANDER J. NEDZEL.

This article will be published in full in a later issue of the ARCHIVES OF PATHOLOGY.

NATURE OF EPISACROILIAC LIPOMA. EMIL RIES.

I described under the name "episacroiliac lipoma" (*Am. J. Obst.* **34**:490, 1937) tumors of the consistency of lipomas of varying sizes and in varying numbers in the tissues over the posterior surface of the sacroiliac joints. A considerable number were associated with pain, sometimes severe. Their frequency was astonishing, and textbooks do not mention them. One thousand unselected persons of both sexes were examined. Of these, 317 had such tumors, and a considerable proportion had associated pain. After removal the tumors ranged in size from that of a hazelnut to that of a hen's egg. They were lobulated and encapsulated by connective tissue. The usual method of examining tumors by microscopic sections demonstrated only fat cells with connective tissue in larger or smaller bands. Blood vessels and nerves were noted in the septums. No inflammatory changes were observed in the connective tissue. From the microscopic examination a diagnosis of lipoma appeared reasonable. However, a different method of investigation revealed a difference between ordinary lipomas and these episacroiliac tumors. When the connective tissue encapsulating an ordinary lipoma is dissected, there remains a solid mass of fat with a center of fat which may have irregular lobes. These lobes always have uninterrupted connection with the center. In the episacroiliac tumors the dissection demonstrated that the apparent mass consisted of many individual fat lobes which were not connected with each other and were held together only by connective tissue. The individual lobes were packed closely together, often pressed against each other so tightly that one was cupped into another adjacent to it. Removal of the connective tissue revealed small fat lobules without a central core and without continuity with each other. There is, therefore, an essential difference in structure between the ordinary lipoma and the so-called sacroiliac lipoma.

FRANCIS D. GUNN, *President*

Regular Monthly Meeting, April 11, 1938

EDWIN F. HIRSCH, *Secretary*

SPINDLE CELL SARCOMA OF THE PROSTATE GLAND. G. J. RUKSTINAT and C. G. WELLER.

A 2 year old boy had difficulty in urination shortly after an infection of the upper part of the respiratory tract. When attempting to void, he strained until a little urine dribbled and his bowels moved. Alternate acid and alkaline therapy had no permanent effect. Two months after the onset of symptoms, rectal examination disclosed a smooth firm mass in the prostate, which extended on the left of the midline to 1 inch (2.5 cm.) below the umbilicus. A cystogram revealed a large filling defect in the lower third of the bladder. There were a few leukocytes, blood cells and albumin in the urine. Tissues from the mass in the bladder were diagnosed as myxosarcoma. The child rapidly grew worse, became uremic and died two and a half months after the onset of symptoms.

The urinary bladder extended 9 cm. above the symphysis pubis and had an infected urachal cyst 3 cm. long and 1 cm. in diameter. The cyst was separated from the bladder by the mucosa of this organ. The bladder was 12 cm. in diameter, and the distal half of the posterior wall was covered, over an area 8 cm. square, with glistening yellow-white gelatinous masses up to 4 cm. in diameter. The largest mass was on a pedicle and was sufficiently close to the urethra to form a ball valve. When these nodules were sectioned, edema fluid escaped, and they collapsed to one third of the original size. The prostate gland was slightly less edematous, pale yellow and 4.2 cm. in diameter. The ureters were twice the normal size, as were the kidneys, and both had an advanced ascending inflammation. There were no metastases in the abdominal viscera.

The growth was a spindle cell sarcoma of the prostate, involving the bladder. The lining of the bladder was intact but raised by edema from the underlying infiltrated muscle. Where the edema was less marked, the cells were spindle shaped, grew in brushlike masses and penetrated the muscle and connective tissue in broad bands. The peripheral parts of the original prostatic tumor and its extensions had the glassy, edematous character but not the stellate structure noted in myxomatous growths.

The patent urachus was infected apparently at the same time as were the bladder, ureters and kidneys. From all these places, colon bacilli, staphylococci and a few nonhemolytic streptococci were cultured. Histologically, the inflammation in these regions seemed of the same intensity and duration.

DISCUSSION

P. DELANEY: The stroma of the prostate is fibrous tissue and smooth muscle. Which tissue formed the sarcoma?

G. J. RUKSTINAT: The growth seems to be fibroblastic. In several places the tumor cells had invaded the muscle proper, and there the two tissues were in sharp contrast.

SUPPURATIVE INTRATHORACIC THYROIDITIS: REPORT OF A CASE. J. D. KIRSHBAUM and A. H. ROSENBAUM.

Although intrathoracic goiter is relatively frequent, suppuration as a complication of it is rare. A review of the literature disclosed only a single case, that of a young woman with a spontaneous abortion. An operation demonstrated suppuration of an intrathoracic goiter, due to gas bacilli. Of 1,985 persons with goiters treated surgically at the Cook County Hospital since 1929, there were only 4 with thyroiditis—an incidence of 0.2 per cent. In 9,888 consecutive autopsies

during the same time, 1,222 nodular goiters were observed, of which 3 were true intrathoracic goiters—an incidence of 0.24 per cent. Hemorrhage, cystic degeneration and other regressive changes are relatively common in substernal goiter, but suppuration is rare.

A Negress, aged 27 years, had been sick at home for eight days. She had lobar pneumonia involving the lower lobe of the right lung. For four weeks she continued to have a fever and became lethargic. A roentgenogram of the chest showed a marked widening of the superior mediastinal shadow. At autopsy, the thyroid gland was markedly enlarged, and from the lower pole of the right lobe extended a lobular mass into the superior and anterior mediastinum. The mass contained numerous nodes and abscesses. Stains revealed gram-positive, lancet-shaped diplococci. The anatomic diagnosis was: multiple abscesses of a nodose intrathoracic goiter, bilateral nodose goiter, compression of the trachea and of the innominate veins by thyroid tissue, slight hypertrophy of the heart, passive congestion of the lungs, acute hyperplasia of the spleen and acute glomerulonephritis. As the retrosternal goiter was adherent to the mediastinal pleura, the infection probably spread to the thyroid by continuity. In suppurative thyroiditis the route of infection is usually hematogenous. Clinically, the main symptoms of substernal goiter are due to compression of the mediastinal structures. With septic manifestations there may be suppurative thyroiditis.

The complete report appears in the May issue of the *Archives of Surgery*, page 867.

A Mixed Tumor of the Salivary Gland Type on the Ankle. I. Davidsohn and R. Bard.

The anatomic designation attached to this group of tumors, on account of their frequency in or near the salivary glands, became incongruous when more and more of these growths were found at sites other than the salivary glands, and particularly since they were discovered on the extremities. Tessman (Inaug. Dissert., Wurzburg, 1911), Kreibig (*Frankfurt. Ztschr. f. Path.* **42**:28, 1931), Hirsch (Arch. Path. **16**:494, 1933), Gaethgens (*Frankfurt. Ztschr. Path.* **47**: 374, 1934) and Scharla (ibid. **49**:269, 1936) reported the first 6 cases. The case reported now is the seventh. A tumor 20 mm. in maximal diameter and located between the left external malleolus and the achilles tendon in a white woman of 50 years was removed Sept. 10, 1937. It was not adherent to the bone and only slightly to the skin. It resembled cartilage. Microscopically, it had the structure of a so-called mixed tumor of the salivary gland type. In addition, it contained some bone tissue. It did not have the typical squamous epithelium and epithelial pearls frequently found in these tumors. Roentgenograms of the bones in the ankle showed no abnormalities. The patient had a recurrence of the growth, which was removed March 26, 1938. This measured 20 by 7 mm. It was adherent to the skin but not to the underlying structures. In gross and microscopic structure it was like the first tumor.

A review of the literature shows a steadily increasing accumulation of observations which favor the concept of an essentially epithelial nature of these tumors despite the morphologic features.

The report will be published in full elsewhere.

Persistent Cloaca and Anal Atresia with Esophageal Atresia and Tracheal Fistula. H. C. Sweany and L. Parmacek.

Persistent cloaca, esophagotracheal fistula, esophageal atresia and anal atresia are combined rarely. A white girl was born after eight months of gestation. The mother's history was negative. The child was well developed and well nourished. The anus was absent. The labia majora and minora were normal. Through an incision at the usual site of the anus, an unsuccessful attempt was made to find the rectum. The infant regurgitated all fluids taken. Exploration of the esophagus with a catheter revealed complete obstruction not far from its upper extremity. Death occurred twenty-four hours after birth. There was no

external urethral opening, for no true vestibule existed. Instead, a deep passage between the labia minora extended upward and posteriorly. Close together in the depths were the openings of the urethra (1 mm. in diameter), the vagina (2 mm.) and the rectum (1 mm.). The urinary bladder, vagina, uterus, tubes and ovaries were normal. The terminal 8 mm. of the rectum was narrowed to 2 mm. in diameter. The colon above was distended with meconium to a diameter of 15 mm. A large subperitoneal hematoma in the right parametric region communicated with the incised wound in the anal region.

The second group of anomalies centered about the trachea and esophagus. The upper third of the esophagus ended in a blind pouch about 1 cm. from the pharyngeal orifice. At the tracheal bifurcation, somewhat posteriorly, the lower esophageal segment began with a 1 mm. opening into the trachea. It widened immediately, however, to the same diameter as the upper esophageal segment and extended to the stomach.

Although anal atresia itself is not uncommon, the primitivity of the cloacal anomaly presented here is unusual, especially when this is combined with esophageal atresia and fistula. The cloaca is the space into which are discharged the urinary, genital and intestinal products. The cloaca is divided by the cloacal septum into the primitive urogenital sinus ventrally and the rectum dorsally. The uterovaginal anlage (fusion of the müllerian ducts) opens high in the median dorsal wall of the deep primitive urogenital sinus. Simultaneously and above this opening, the vesicourethral anlage opens into the urogenital sinus. The urogenital sinus becomes shallower as the urethral and vaginal openings descend toward the exterior. Finally, both these orifices, through the growth of the urethrovaginal septum, open separately into the shallow final remnant of the urogenital sinus, the vestibule.

In embryologic development, the laryngotracheal groove forms on the anterior wall of the esophagus, caudal to the pharynx. Two furrows deepen longitudinally along the sides of the respiratory anlage until they separate it from the esophagus. Incomplete separation explains the esophagotracheal fistula. Just above the fistula, the temporary epithelial esophageal narrowing, due to epithelial overgrowth, becomes permanent. Lewis mentioned the presence of two lateral folds in the embryonal esophagus at the level of the tracheal bifurcation. These may fuse and cause stenosis of the upper esophageal segment. Aschoff mentioned the possibility of the development of the lateral furrows too far dorsally and cranially as a factor in esophageal atresia with tracheal fistula.

From a review of the embryology of the cloaca and the tracheoesophageal region it appears that the anomalies of this case are due to an arrest of development in a primitive phase. The result is a persistent cloaca and atresia of the esophagus with esophagotracheal fistula.

BUFFALO PATHOLOGICAL SOCIETY

ERNEST WITEBSKY, *President*

Regular Meeting, March 26, 1938

SAMUEL SANES, *Secretary*

THE RENAL LESION INDUCED BY MERCURY. J. GRAHAM EDWARDS.

The kidneys of persons dying from mercury poisoning and those of rabbits, guinea pigs, rats and frogs dying or killed at various intervals after similar poisoning have been studied with special reference to the site and character of the lesion within the tubule. In the mammalian kidney, except that of man and that of the guinea pig, the most commonly involved site, showing complete necrosis, when

the animal has been subjected to intraperitoneal injection of 0.3 mg. of mercury bichloride per hundred grams of body weight, is the third fourth (from 2 to 3 mm.) of the proximal convolution. In man the first three fourths of this convolution is totally necrotic (effective dose unknown), and in the kidney of the guinea pig the lesion involves the terminal fourth and the thin segment of the medullary loop of Henle. In the kidneys of the other mammals studied, including man, this terminal fourth appears histologically normal even when fairly often the first three fourths (beginning at the renal corpuscle) is partially or, in some instances, entirely necrotic.

When the frog receives via the dorsal lymph sac an injection of 1.6 mg. of mercury bichloride per hundred grams of body weight, the resulting renal lesion is restricted to the proximal convolution and commonly to its middle third. In about equal numbers of animals, however, the lesion involves the lower half or three fourths and sometimes the entire convolution. In both the frog and the mammal the parts of this convolution most resistant to mercury bichloride are generally the initial and the terminal portion, although in the frog the initial portion is more resistant to the action of mercury than the terminal—a condition opposite to that observed in man, the rabbit and the rat.

The lesion resulting from the action of mercury is definitely restricted to the proximal convolution in the kidney of the frog and that of the mammal. It is usually sharply segmental within this convolution, making the kidney appear in stained sections as though some tubules were necrotic and some were not, a condition which rarely occurs. Injury to other parts of the tubule is more apparent than real and only occasionally apparent.

PROLONGED DIABETES MELLITUS CULMINATING IN VON GIERKE'S DISEASE IN A YOUTH OF CHILDLIKE HABITUS. K. TERPLAN, S. VOGEL and E. HYDE.

A white American boy, 15 years of age, was known to be diabetic for nine years and during this time was treated with insulin, 30 units daily. On admission to the hospital because of acute gastroenteritis, he was described as distinctly underdeveloped, having the appearance of a child of about 6 years. The genitalia were entirely infantile: no secondary sex characteristics were developed. The urine contained considerable sugar. The blood dextrose was 216 mg. per hundred cubic centimeters but later decreased to 125 mg. At this time, about one half year before the death of this patient, the edge of the liver was palpable 3 fingerbreadths below the costal margin. The patient was discharged, improved. On second admission, four days previous to death, he was in coma, a condition which had been preceded by a cold. The blood dextrose amounted to 69 mg. per hundred cubic centimeters (most probably under the effect of insulin); the amount later rose to 286 mg. and then, on the day of death, to 330 mg. On admission the urine showed dextrose (4 plus) and on the last day became free from dextrose. The clinical diagnosis throughout the course of the disease was diabetes mellitus and infantilism. At the second admission the liver was found to be tremendously enlarged and the spleen palpable.

The typical picture of von Gierke's disease of the liver was found post mortem: The liver, which almost filled the abdomen, weighed 2,550 Gm. Careful study, together with chemical analysis, revealed marked storage of glycogen (the glycogen content of the liver about 10 per cent) and fatty changes. Of the other tissues analyzed for glycogen content—heart, striated muscle and kidney—only the kidney showed a pathologic content of glycogen. Droplets and granules were found in the loops of Henle and in some collecting tubules; the amount, however, did not exceed that seen in severe diabetes mellitus. The pancreas was markedly atrophic (weight, 21.5 Gm.). Histologically, the observations in the pancreas pointed to true juvenile diabetes, with extreme reduction in the number of the islands of Langerhans; those few which were present, appeared fairly normal or slightly hypertrophic. The testicles, epididymes, seminal vesicles and prostate were entirely of infantile structure. The pituitary, adrenals, thyroid and parathyroids were grossly and histologically normal.

This case presents, in our opinion, a combination of true juvenile diabetes with infantilism and typical von Gierke's disease of the liver. The literature as far as we have been able to discover contains reports of but 2 cases of like nature; these were described by Pierre Mauriac in a clinical study. Diabetes mellitus is frequently described as combined with various endocrine disorders, especially those of delayed growth and hypoplastic constitution, in many cases ascribed to disorders of the pituitary function, but the combination of diabetes, infantilism and storage of glycogen in the liver is apparently more rare.

From the history in this case it is certain that the enlargement of the liver was noticed only in the last year, while typical diabetes, which was controlled by insulin and diet, was present for nine years. It is suggested that the prolonged administration of insulin might be one pathogenic factor in this case responsible for the pathologic storage of glycogen in the liver.

FURTHER OBSERVATIONS ON PRIMARY SERUM TOXICITY AS DEMONSTRATED BY MEANS OF THE CHICKEN EMBRYO. E. WITEBSKY and E. NETER.

It was previously reported (Baumann, A., and Witebsky, E.: *Ann. Inst. Pasteur* **53**:282, 1934. Witebsky, E., and Neter, E.: *J. Exper. Med.* **61**:489, 1935) that serum containing Forssman antibodies when dropped on the surface of a chicken embryo may induce the following changes: The vascular network shrinks, the body of the embryo sinks into the depth of the yolk, and the embryo finally dies. The potency of the serum to produce this phenomenon is destroyed on inactivation of the serum. On the other hand, it is possible to restore this activity in an inactivated Forssman antiserum by the addition of complement. Meanwhile, it was found that Forssman antiserums from different sources, such as a blood group A antiserum, were as potent as sheep cell antiserum. It appeared to be of interest to examine the serum of patients suffering from infectious mononucleosis as to its capacity to induce the phenomenon under investigation. When dropped on the surface of a chicken embryo, this serum behaved exactly as did normal human serum. It induced the phenomenon in dilutions up to 1:5 and could not be reactivated by the addition of guinea pig serum. Thus antibodies present in the serum of patients with infectious mononucleosis seem to be different from the Forssman antibodies produced by immunization.

Two substances were found which when added to potent serums inhibited the reaction in the chicken embryo. One was tannic acid (a neutralized 1 per cent solution) and the other sodium polyanetosulfonate (used in the form of liquoide Roche). While the effectiveness of the latter preparation is probably due to the inactivation of complement, the action of tannic acid seems to be more complex.

Moccasin venom when dropped on a chicken embryo was capable of producing severe hemorrhages by virtue of the hemorrhagin present in this type of venom. When tannic acid was mixed with this venom, its hemorrhagic effect toward the chicken embryo was neutralized. These observations parallel those of Freund (*J. Immunol.* **21**:127, 1937), who found that diphtheria and tetanus toxin may be neutralized by tannic acid, and of Klopstock and Neter (*Biochem. Ztschr.* **261**:207, 1933), who showed that tannic acid may neutralize the toxic and lethal action of cobra venom toward mice.

ANOMALOUS DRAINAGE OF PULMONARY VEINS INTO THE CORONARY SINUS. S. SANES and K. TERPLAN.

Up to the present the available literature yields only 3 cases of anomalous drainage of all the pulmonary veins into the coronary sinus as a primary abnormality of development (Nbarro; De Groat and Thatcher; Nagel).

A white boy, 5 months and 19 days of age, was admitted to the hospital because of edema of the legs and frequent emesis. There was no history of cyanosis. Physical examination revealed anemia. The heart was not enlarged. No thrills or murmurs were detected. The liver was palpable. Edema of the legs was present. Death occurred in a cyanotic attack.

At autopsy the heart was markedly enlarged. This enlargement consisted of the right atrium, the right auricular appendage and the right ventricle. The left atrium and ventricle were small and were rotated posteriorly. The positions of the orifices of the superior and inferior venae cavae were normal. The valve of the inferior vena cava was well developed and continued with a thick limbus fossa ovalis. The fossa ovalis was deep and showed a small foramen ovale. The opening of the coronary sinus was enlarged; it measured 1.5 cm. in circumference. The coronary sinus itself showed marked dilatation and thickening of the wall. Into it opened a right and a left pulmonary venous trunk, each of which was formed by veins from the respective lobes. In addition, the sinus received the great cardiac vein, the middle cardiac vein, the oblique vein and two atrial veins. The right ventricle was dilated and hypertrophic. The pulmonary artery was increased in circumference. Grossly, the ductus arteriosus could not be probed.

A review of 3 reported cases along with our own discloses such a uniformity of anatomic observations as to classify anomalous drainage of the pulmonary veins into the coronary sinus among the specific cardiac malformations. The anomaly appears most satisfactorily explained on the basis of an abnormal connection in embryonic life between the pulmonary venous trunk and that portion of the sinus venosus which develops into the coronary sinus.

Books Received

RECENT ADVANCES IN PATHOLOGY. Geoffrey Hadfield, M.D., F.R.C.P. (London), Professor of Pathology, University of London, and Pathologist to St. Bartholomew's Hospital, London; and Lawrence P. Garrod, M.A., M.D., B.Ch. (Cambridge), F.R.C.P. (London), Professor of Bacteriology, University of London, Bacteriologist to St. Bartholomew's Hospital, and Examiner in Pathology, University of Cambridge. Third edition. Cloth. Price, $5. Pp. 420, with 65 illustrations. Philadelphia: P. Blakiston's Son & Co., Inc., 1938.

INTRODUCTION TO OPHTHALMOLOGY. Peter C. Kronfeld, M.D., Professor of Ophthalmology, Peiping Union Medical College. Cloth. Price, $3.50. Pp. 331, with 44 illustrations. Springfield, Ill.: Charles C. Thomas, Publisher, 1938.

CLINICAL PATHOLOGY. Edited by Roy R. Kracke, B.S., M.D., Professor of Pathology, Bacteriology and Laboratory Diagnosis; Chairman of the Department, Emory University. Fabrikoid. Price, $6. Fp. 582, with 236 illustrations. Baltimore: William Wood & Company, 1938.

A CRITICAL INVESTIGATION OF THE BLOOD GROUPS AND THEIR MEDICO-LEGAL APPLICATION. Dawood Matta, M.B., Ch.B. (Cairo), Ph.D. (Glasgow), Lecturer of Forensic Medicine, Faculty of Medicine, University of Egypt, Cairo. Pp. 231. Cairo: The Egyptian University, 1937.

LA RÉACTION DE FIXATION DANS LES TUBERCULOSES. HUMAINES ET ANIMALES. Achille Urbain. Second edition. Paper. Price, 28 francs. Pp. 146. Paris: Masson & Cie, 1938.

CORRECTIONS

In the article by Drs. Steinberg and Martin, entitled "Inflammation of Serous Surfaces: Factors Modifying Types of Cell Response," in the June issue (ARCH. PATH. **25**:792, 1938), the second sentence in the second paragraph under "Comment," on page 800, is a repetition and should be deleted.

In the last line of Dr. Paul Klemperer's discussion of the paper by Dr. N. Chandler Foot, entitled "Epidermoid Carcinoma of the Breast," on page 768 of the May issue of the ARCHIVES, the word "intracellular" should be "intercellular."

THE ARCHIVES OF PATHOLOGY is published by the American Medical Association as a medium to advance pathology in the United States and to promote research and observation in this field.

Manuscripts for publication, books for review and correspondence relating to contributions should be sent to Dr. Ludvig Hektoen, Chief Editor, 535 South Wood Street, Chicago, or to any other member of the Editorial Board. Communications regarding subscriptions, reprints, etc., should be addressed Archives of Pathology, American Medical Association, 535 North Dearborn Street, Chicago.

Articles are accepted for publication on condition that they are contributed solely to the ARCHIVES OF PATHOLOGY. Manuscripts must be typewritten, preferably double spaced, and the original copy should be submitted. Zinc etchings and halftones will be supplied by the Association when the original illustrations warrant reproduction and when their number is not considered excessive.

Footnotes and bibliographies (the latter are used only in exhaustive reviews of the literature) should conform to the style of the Quarterly Cumulative Index Medicus. This requires, in the order given: name of author, title of article and name of periodical, with volume, page, month and day of month if the journal appears weekly—and year.

Matter appearing in the ARCHIVES OF PATHOLOGY is covered by copyright, but as a rule no objection will be made to its reproduction in a responsible medical journal if proper credit is given. However, the reproduction for commercial purposes of articles appearing in the ARCHIVES OF PATHOLOGY or of any of the other publications issued by the Association will not be permitted.

The ARCHIVES OF PATHOLOGY is published monthly. The annual subscription price (for two volumes) is as follows: domestic, $6.00; Canadian, $6.50; foreign, $7.00, including postage. Single copies, except the July 1938 issue, 75 cents, postpaid; the July issue, $2.00.

Checks, money orders and drafts should be made payable to the American Medical Association.

OTHER PERIODICAL PUBLICATIONS
of the American Medical Association

THE JOURNAL OF THE AMERICAN MEDICAL ASSOCIATION—Weekly. Carries all the medical editorials and matters of general medical interest. Illustrated. Annual subscription price (two volumes): domestic, $7.00; Canadian, $8.00; foreign, $11.00. Single copies, 25 cents.

ARCHIVES OF INTERNAL MEDICINE—Monthly. Devoted to the publication of advanced original clinical and laboratory investigations in internal medicine. Illustrated. Annual subscription price (two volumes): domestic, $6.00; Canadian, $6.50; foreign, $7.00. Single copies, 75 cents.

AMERICAN JOURNAL OF DISEASES OF CHILDREN—Monthly. Presents pediatrics as a medical science and as a social problem. Includes carefully prepared reviews based on recent pediatric literature; abstracts from foreign and domestic literature, book reviews, transactions of special societies, etc. Illustrated. Annual subscription price (two volumes): domestic, $6.00; Canadian, $6.50; foreign, $7.00. Single copies, 75 cents.

ARCHIVES OF NEUROLOGY AND PSYCHIATRY—Monthly. A medium for the presentation of original articles on nervous and mental diseases, with abstracts from foreign and domestic literature, book reviews, transactions of special societies, etc. Illustrated. Annual subscription price (two volumes): domestic, $6.00; Canadian, $6.50; foreign, $7.00. Single copies, 75 cents.

ARCHIVES OF DERMATOLOGY AND SYPHILOLOGY—Monthly. Devoted to advancing the knowledge of and progress in cutaneous diseases and syphilis. Publishes original contributions and abstracts of the literature on these two subjects, transactions of important dermatologic societies, book reviews, etc. Illustrated. Annual subscription price (two volumes): domestic, $6.00; Canadian, $6.50; foreign, $7.00. Single copies, 75 cents.

ARCHIVES OF SURGERY—Monthly. Devoted largely to the investigative and clinical phases of surgery, with monthly reviews on gastroenterologic and urologic surgery. Well illustrated. Annual subscription price (two volumes): domestic, $6.00; Canadian, $6.50; foreign, $7.00. Single copies, 75 cents.

ARCHIVES OF OTOLARYNGOLOGY—Monthly. A medium for the presentation of original articles on diseases of the ear, nose and throat, with abstracts from foreign and domestic literature, book reviews, transactions of special societies, etc. Illustrated. Annual subscription price (two volumes): domestic, $6.00; Canadian, $6.50; foreign, $7.00. Single copies, 75 cents.

ARCHIVES OF OPHTHALMOLOGY—Monthly. Includes original articles on diseases of the eye, abstracts from foreign and domestic literature, book reviews, transactions of special societies, etc. Illustrated. Annual subscription price (two volumes): domestic, $6.00; Canadian, $6.50; foreign, $7.00. Single copies, 75 cents.

QUARTERLY CUMULATIVE INDEX MEDICUS—Quarterly. A complete subject and author finder to the worth-while current medical literature of the world. Issued four times a year. Second and fourth volumes bound for permanent reference. Subscription price, calendar year: domestic, $12.00; Canadian, $14.00; foreign, $14.00.

AMERICAN MEDICAL ASSOCIATION
535 North Dearborn Street CHICAGO